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ABSTRACT

This workbook contains laboratory exercises designed for use in a college introductory biology course. Each exercise helps the student develop a basic science skill. The exercises are arranged in a hierarchical sequence suggesting the scientific method. Each skill facilitates the development of succeeding ones. Activities include Use of the Microscope, Cell Types, DNA Replication, and Inheritance of Genetic Traits. Process skills such as observing, integrating, classifying and predicting are emphasized. Most exercises are structured to fit into a two-hour laboratory period. Each exercise provides general objectives, behavioral objectives, introduction, materials and equipment, procedure, references, and report sheet. (MA)

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# LABORATORY ACTIVITIES FOR DEVELOPING PROCESS SKILLS

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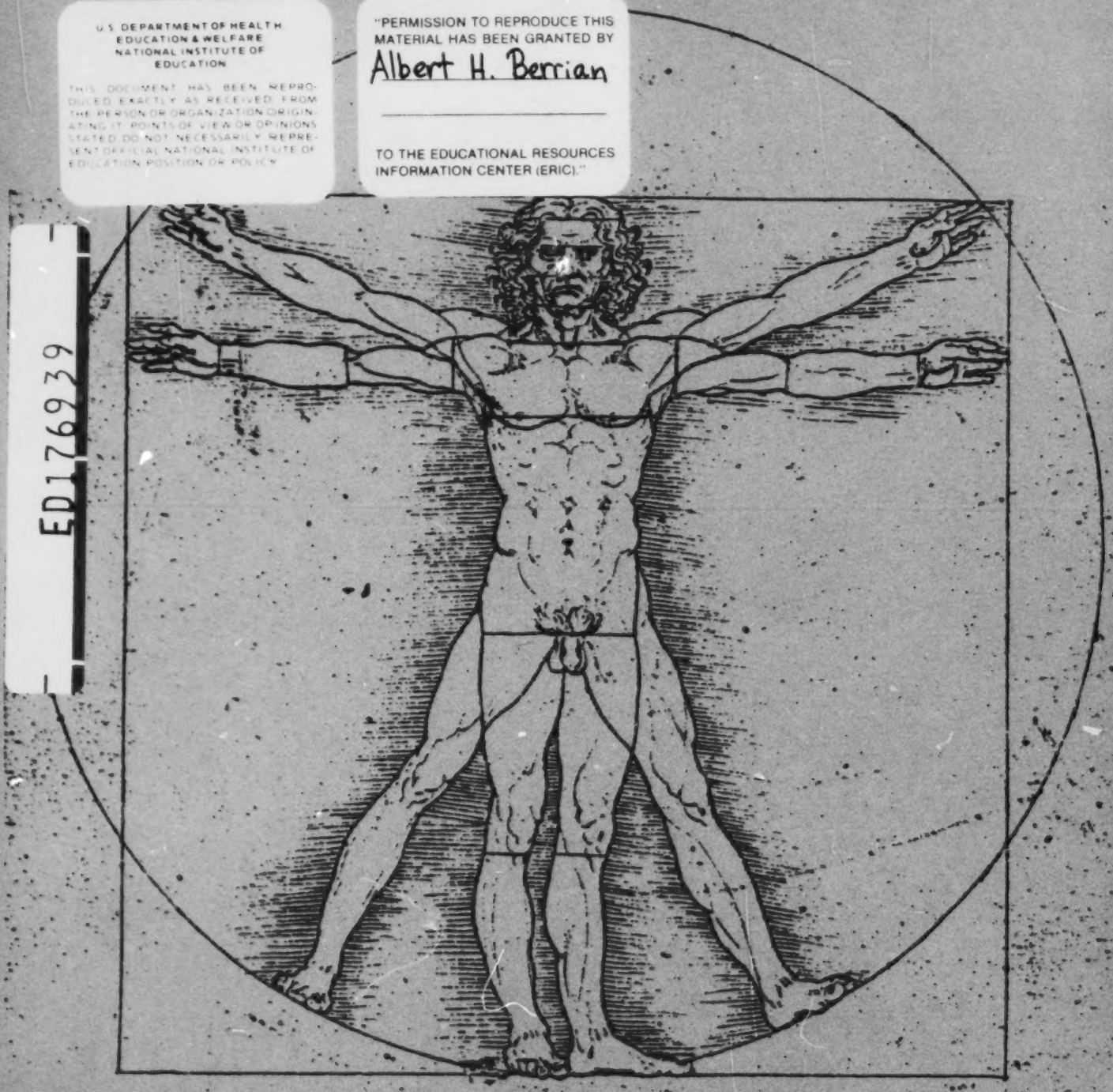

SE 027 569

# LABORATORY ACTIVITIES

# FOR

# DEVELOPING

# PROCESS SKILLS

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# PREFACE

This arrangement of laboratory exercises is designed for use in Introductory Biology Courses for majors and non-majors. It is intended as an aid to the teacher and the student in developing basic Science Skills which should have broad application.

The exercises are arranged in a hierarchical sequence which suggests how a scientist might work. The sequence is designed to develop skills needed to cope with the contemporary environment. The development of one skill facilitates the development of succeeding skills. These skills are listed below:

Exercise 1.1 - Observation

Exercise 1.2 - Microscope Observation

Exercise 2 - Using Numbers

Exercise 3.1 - Using Numbers

Exercise 3.2 - Integrating

Exercise 4 - Using Space-Time Relationships

Exercise 5 - Communicating

Exercise 6 - Inferring

Exercise 7 - Classification

Exercise 8 - Communicating

Exercise 9 - Predicting

Exercise 10 - Predicting

Exercise 11 - Space-Time Relationships

Exercise 12 - Integrating

Exercise 13 - Integrating

Exercise 14 - Space-Time Relationships

Most of the exercises are designed to suit two-hour laboratory periods, while others are structured to permit a break in time for the students and to facilitate the division of an exercise into independent parts. In general, the exercises include extra material to allow the teacher an opportunity for broader selection based upon student ability and existing college facilities.

Each exercise includes general objectives and more specific behavioral objectives to aid in the determination of the student's progress. There is an introductory section before each exercise. The introduction and references are intended to direct and assist the student to expand his knowledge through the use of the library.

The exercises provide instructors with an opportunity to guide the students in actively solving problems efficiently and analytically. They provide for active participation by each student, and require students to master the basic tools of biology needed for efficient problem-solving techniques.

## ACKNOWLEDGEMENTS

The arrangement and modification of exercises found in this package represent the efforts of the interested and dedicated teachers who attended a Summer Conference at Dillard University in New Orleans, Louisiana in 1978. The teachers involved in the task were:

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## TABLE OF CONTENTS


PREFACE

i - ii

ACKNOWLEDGEMENTS

iii

| EXERCISE | RELATED DISCUSSION UNIT | | PAGE NO. |
|---|---|---|---|
| 1.1 | 1 | HOW KNOWLEDGE IS ACQUIRED - THE BLACK BOX | 1 |
| 1.2 | 1, 2 | USE OF THE MICROSCOPE | 7 |
| 2. | 1 | CONCEPTS OF MASS, WEIGHT AND DENSITY | 22 |
| 3.1 | 1 | TOOLS FOR THE STUDY OF BIOLOGY | 34 |
| 3.2 | 2, 3, 7 | CELL TYPES | 41 |
| 4. | 3 | DIFFUSION, OSMOSIS AND ACTIVE EXCLUSION | 50 |
| 5. | 6 | WATER CONTENT OF VARIOUS TISSUES AND CELLS | 62 |
| 6. | 3 | COACERVATES AND EMULSIONS | 66 |

| | | | |
|---|---|---|---|
| 7. | 2, 7 | THE PHYLA OF THE ANIMAL AND PLANT KINGDOMS | 71 |
| 8. | 3, 5 | DNA REPLICATION AND TRANSCRIPTION BY $_{m}$RNA AND $_{t}$RNA TO SEQUENCE A PROTEIN | 77 |
| 9. | 5 | THE INHERITANCE OF GENETIC TRAITS | 84 |
| 10. | 5, 6 | THE INHERITANCE OF SPECIFIC PROTEINS | 97 |
| 11. | 5 | DO ENVIRONMENTAL FACTORS AFFECT THE ACTION OF GENES? | 107 |
| 12 | 6 | INFLUENCE OF THYROID HORMONE ON RATE OF DEVELOPMENT | 112 |
| 13 | 6, 8 | EFFECT OF PHYSICAL AND CHEMICAL FACTORS ON ANIMALS | 116 |
| 14 | 8 | GROWTH OF A POPULATION | 123 |


Cover Design: The Proportions of the Human Figure, Leonardo da Vinci. c. 1492

# EXERCISE 1.1 - HOW KNOWLEDGE IS ACQUIRED - THE BLACK BOX

## GENERAL OBJECTIVE:

To develop an appreciation for the processes of observation and inference that scientists generally use in formulating theories.

## BEHAVIORAL OBJECTIVES:

Upon completion of this exercise, the student should be able to:

1. Specify the things that he did in conducting the black box experiment, which coincided with the observation, the problem, the hypothesis, the experiment and the theory of the Scientific Method.
2. Indicate the aims and objectives of Science.
3. Indicate the limitations of the Scientific Method.
4. Indicate the significance of the control in an experiment.
5. Indicate control in the black box experiment.
6. Indicate specific instances in the black box experiment in which the skill of inference is used.

## INTRODUCTION

The scientific method as used in biology is a testing procedure which has come to be accepted by scientists. It reached its present state after scientists in different centuries found that its several components were necessary in order to take concepts and philosophies and use them to solve both practical and theoretical problems. Therefore, a scientific experiment is an experience in applied philosophy.

Scientific reports in biology reflect the use of the scientific method. They are usually written in five sections, and scholars and researchers have come to look in specific sections for the kind of information they seek.

## PROCEDURE

Part A

You will be provided with a closed box of small size containing an object. Your test is to determine as many facts about the object in the box as you can and to finally guess what is in the box. A set of objects will be available, some, all, or none of which may be in boxes given to members of the class. There will also be some balances and an empty box which looks like yours. Write down observations and the procedures that you use as you go along, and also your conclusion as to the contents. The teacher will let you know when you can open the box. Were you right? If you were wrong, how do you feel about it? If you were wrong, obtain a second box and see if you can do any better. Relate what you did to the steps in the scientific method.

### Part B. Scientific Reports (Original Papers)

The first section is called INTRODUCTION, and gives the historical precedents for doing the experiment or experiments. The introduction ends with the hypotheses or objectives of the experiments. It then follows that the historical part should marshal evidence that the objectives are reasonable.

The second section is called MATERIALS AND METHODS. In this section are listed the materials used, specific organs or tissues used, the organism from which they came, and the procedure used. (This section may come from the laboratory instructions so that they may be included as they are, but with any additional items properly entered.).

The third section is called RESULTS. In this section report your laboratory data and the requested computations (as a minimum) based thereon. In order to have results to report, it will be necessary to keep a laboratory notebook. The notebook is required of all students. Complete your data and computations in your notebook.

The REPORT SHEETS for the exercises in this workbook constitute a kind of notebook.

The fourth section is called DISCUSSION. The fundamental aim here is to explain what happened in the experiment. This process of explanation will involve looking up information so that you can tell whether or not your results are in line with the findings of others. The chief source of this

information is to be found in original papers appearing in the scientific journals, reference treatises and sometimes even in textbooks and manuals. The best way to begin looking up such information is to go to an abstract journal such as Chemical Abstracts or Biological Abstracts. Look up the subject in the annual indices, starting with the most recent and working backward through the years. Index references in Chemical Abstracts are listed by number. Biological abstracts are listed by abstract numbers. In both cases abstracts are grouped by category, but many times they may be in some other category also. Because of this, the abstract will be cross-referenced by several subjects, animals, organs, or procedures. The abstract is an abbreviated summary of an original article. By reading it you may either obtain the desired information or you may determine whether or not you need to read the original paper. The standard form for references is given in the next section. Other primary sources are the Annual Review of Physiology, Annual Review of Biochemistry, and others in the Annual Review series. Advances in Carbohydrate Chemistry, Advances in Colloid Chemistry, and Recent Advances in Hormone Research are titles of other annual series which seek to review recent developments. A list of journals relevant to this course will be found on the laboratory bulletin board.

The fifth section of the report is entitled SUMMARY AND CONCLUSIONS. This is a short section which closes the report. Note that this section has two parts. The summary precedes the conclusions based on your data.

The last part of the report is a list of REFERENCES used. The term "Bibliography" is used only for complete listings of books and papers on a given subject and therefore will not apply here. References may be made several ways in the text of the report. We shall consider it standard in the course to list the names of the author or authors in the same order in which they appear under the title of the original paper, followed by the year of publication. If the same sequence of authors has more than one publication add a small a, b, c, etc. after the year. In the reference list give the last name of the first author followed by his/her initials. If there is more than one author, those after the first are listed with their initials appearing first. After this comes the year of publication, then the title of the paper, the journal, the volume and page numbers.

EXAMPLES:

> Clausen, H. H., 1940. "The atrophy of the adrenal cortex following the administration of large amounts of progesterone". Endocrinology, 27:989-993.

Gaunt, R. G., C. H. Tuthill, N. Antochank and J. H. Leathem., 1953. "Antagonists to cortisone: An ACTH-like action of steroids". Endocrinology. 54:273-283.

Here is an example of a book reference:

Cohen, G. N., 1969. Cell Regulation. Holt, Rinehart and Winston, Inc. Coldspring Harbor Symposia on Quantitative Biology, 26, 1961 Cellular Regulatory Mechanisms. Coldspring Harbor, L.I., New York.

## REFERENCES


Aaron, A. B. and A. M. Bork, 1964. Science and Ideas, Prentice-Hall, Englewood Cliffs, N.J.

Baker, J.J.W. and G.E. Allen, 1968. Hypothesis, Prediction, Implication in Biology, Addison Wesley, Reading, Mass.

Beveridge, W. I. B. 1960. The Art of Scientific Investigations, Random House, Inc., New York.

Conant, J. B. 1947. On Understanding Science, Yale University Press, New Haven, Conn.

## REPORT SHEET FOR EXERCISE 1

Notes on the Black Box Experiment:

What do you think is in the box (before you open it)?

Were you right?

How do you feel about it?

Do you think that you should replace the object with one that, in your opinion, should have been there if you were wrong?

Are you going to try the experiment again with another unknown box? If so, use another sheet of paper for your notes and include it here.

Write up a report on your theory as to what is in the black box, using the format suggested in Section B.

## EXERCISE 1.2 - USE OF THE MICROSCOPE

### GENERAL OBJECTIVE:

To learn to use and care for the microscope.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students should be able to:

1. Identify the parts of a compound microscope or stereoscope when given a diagram or the actual microscope.
2. Discuss the uses and limitations of the microscope.
3. Discuss how compound microscopes work.
4. Handle and use the compound and zoom microscopes.

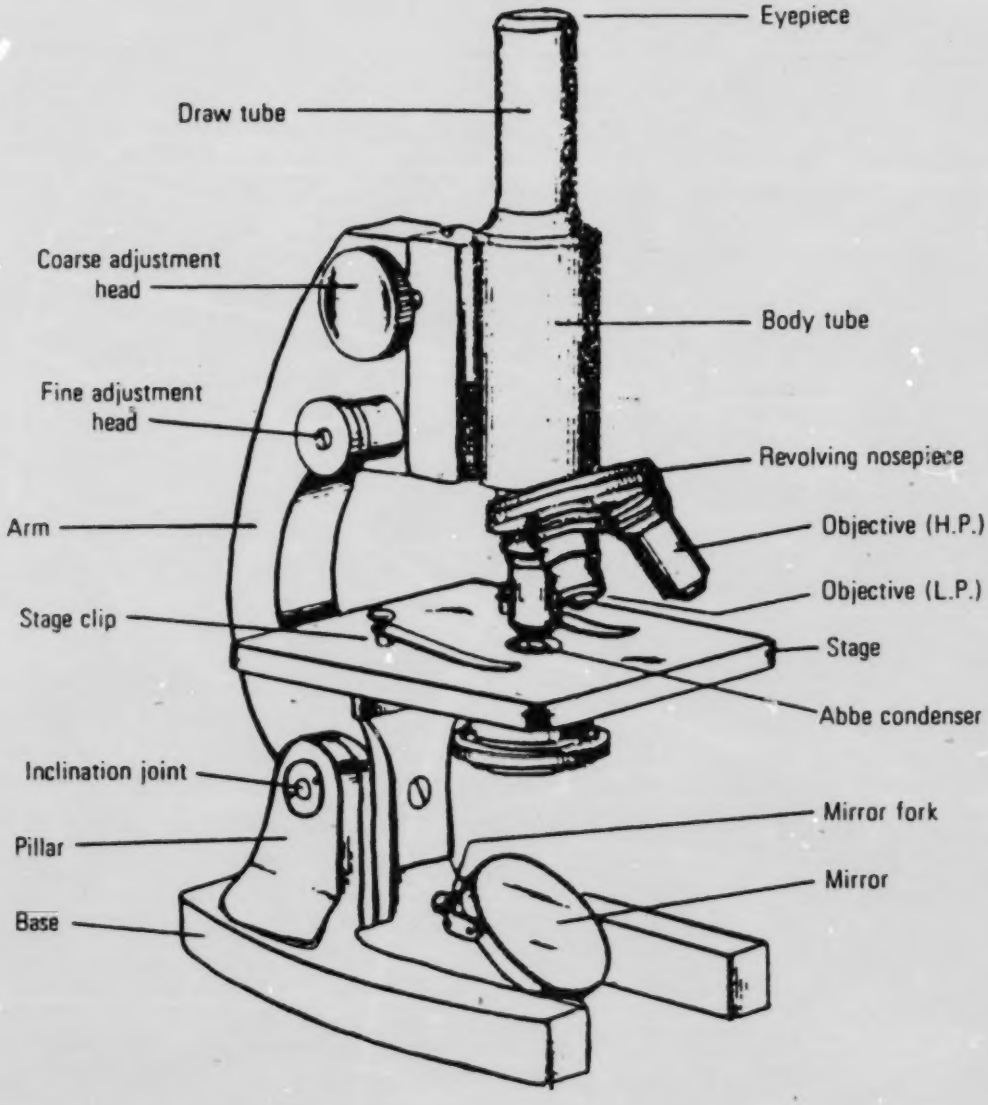
### INTRODUCTION

The microscope is an important optical tool which makes visible details of both biological and physical structures which otherwise cannot be discerned with the naked eye. Microscopes are very costly equipment. Proper handling assures good service and reduces the probability of damage.

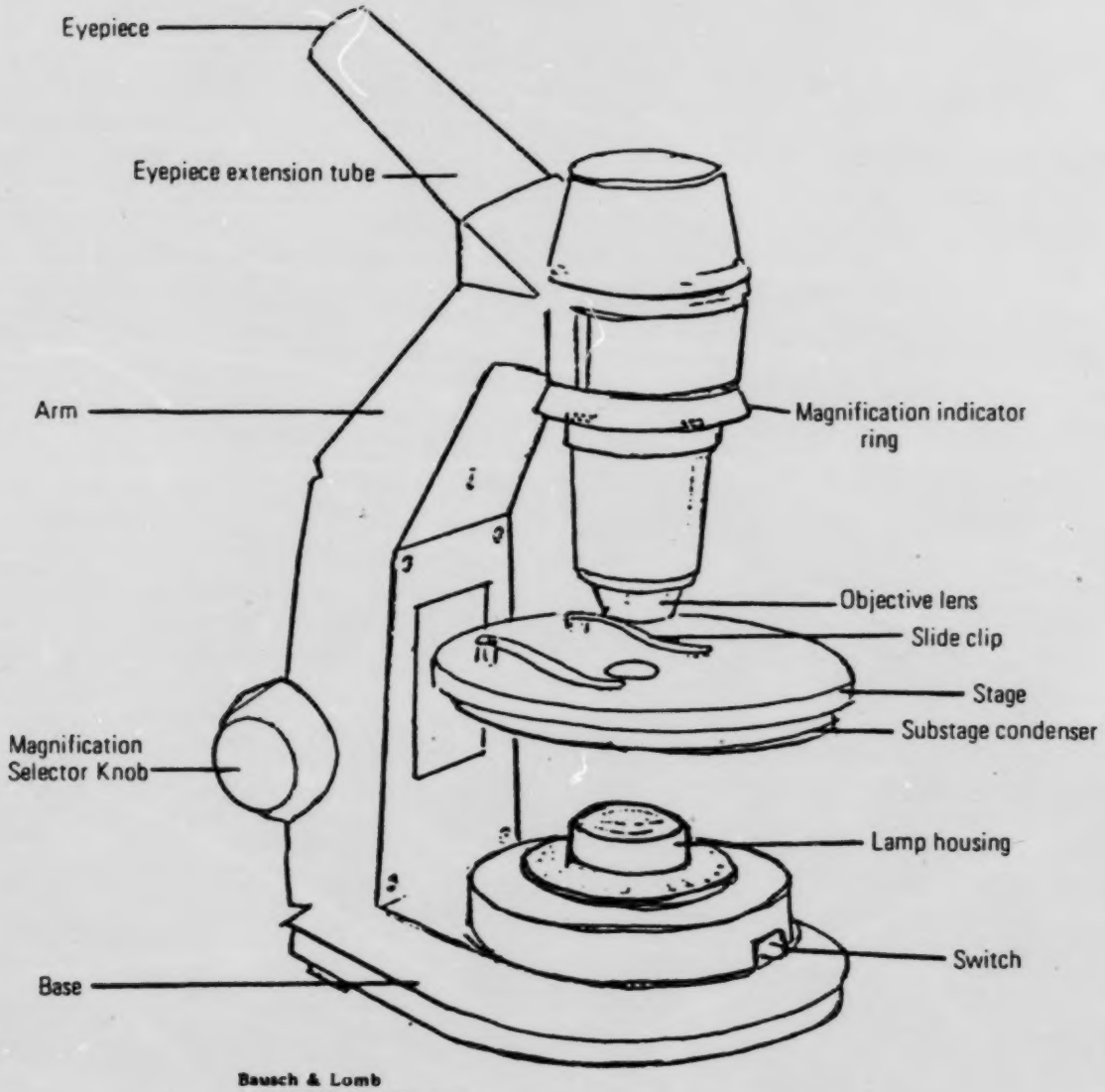
### MATERIALS AND EQUIPMENT

Different types of microscopes
Lens paper
Millimeter rulers
Slides with letter "e"
Clear slides
Cover slips
Pond water
Tissue slides

## Parts of the Compound Microscope




Courtesy of the Bausch and Lomb Optical Company




**Bausch & Lomb**
**ACADEMIC 255 Student**
**Zoom Microscope MODEL 011**

## PROCEDURE

### Part A. Parts of the Compound Microscope

Study the diagram on page 8 and identify the eyepiece, body tube, arm, objectives, condenser, base, stage, etc.

Now read Part B, "Transporting the microscope", by the time you have finished, the teacher will ask you to identify the parts mentioned on a real microscope. You will then be asked to remove the microscope assigned to you from the cabinet to your desk.

### Part B. Transporting the Microscope

The microscope looks heavy and many of them are. This feature is for steadiness when the instrument is in use, and does not mean that the instrument is "rugged" in the usual sense.

1. The microscope must not be jarred. It should not be dropped on the floor or the table top. This will either knock the lenses out of alignment or break them.

2. Pick up the microscope by the arm (never by the body tube) and support the base with your other hand.

3. Carry the microscope upright. Most microscopes have loose eyepieces which will fall out if the instrument is carried in a slant position.

4. Place the microscope upon the table gently.

Now complete the identification of the parts of the microscope making use of the diagram on page 20, and answer the questions on the report sheets.

### Part C. Cleaning the Microscope

1. Always use clean lens paper or optical tissue. Never use handkerchiefs, facial or bathroom tissue, paper towels, typing paper or emery cloth.

2. Wipe the glass surface, changing the place on the lens paper after each wipe. This is done so that if any grit is picked up it is not scrubbed back and forth over the lens. If there is an encrusted

substance on the lens, wet a piece of lens paper with water and wash off the lens, then dry it with lens paper. Do not huff (blow) on the lens to moisten it because small droplets of saliva come out with the breath. The protein in the saliva tends to leave smears on the glass. Clean the following surfaces:

Upper surface of the
eyepiece
Objective lenses
Upper surface of the
condenser mirror

Part D. Making an Observation

1. Rotate the low power objective (marked 1.6 cm) into position. A spring clip seats the nosepiece exactly so that the lenses are in line with the eyepiece. Use the coarse adjustment head to bring the bottom of the objective to about 1.6 cm. (1/2 inch) above the stage.

2. If your microscope has a condenser, adjust the plain side of the mirror so that light is directed through the condenser. If your instrument does not have a condenser, use the concave side of the mirror in order to focus light at the level of the stage.

3. Raise the condenser to stage level. Look through the eyepiece and adjust the iris diaphragm on the condenser or under the stage so that the lighted field is about the same size as the visible field.

4. Place a slide containing a 2 mm. printed scale or a slide with letter "e" on the stage and secure it with the spring clips on stage. Place the slide so that the part to be observed is in the light. Adjust the focus with the coarse adjustment head so that the object is distinctly visible. How does the image move in the field compared with the way that the slide is actually moved on the stage? (Answer on the report sheet.)

5. Measure the diameter of the field to the nearest millimeter (mm). Convert the mm to microns by multiplying by 1,000 (1,000 microns = 1 mm).

6. Compute the magnification of the low power system by multiplying the rated magnification of the ocular (usually it is 10X, but check yours to be sure) by the rated magnification of the low power objective (usually 10X, but check again to be sure).

7. Now rotate the nosepiece to bring the high-dry objective (4 mm) into place. The high-dry objective is usually rated about 45X and is marked for working about 4 mm above the slide. Focus upward with the fine adjustment head. If the scale does not come into focus, your microscope is not parfocal (if it does, it is). There is nothing you can do about this now. To find the object, watch from the side and lower the objective to a position near the slide (2-3 mm above it), but without touching it. Now look through the eyepiece and focus upward with the fine adjustment head. The coarse adjustment head should not be used for focusing high-dry and oil immersion objectives. Adjust field of view. High-dry and oil immersion require increasing amounts of light. Why?

   It is good technique to keep both eyes open. With a little practice you can mentally repress the vision in the eye not being used for microscopy, but at the same time you avoid the strain of keeping your eyelid muscles contracted for long periods of time resulting in muscle fatigue and possibly a little pain.

8. Measure the width of the field, estimating the last fraction of a mm. Convert your measurements to microns and record them on the report sheet.

An Alternate Method for Computing the Diameter of High-Power Field

Divide the magnifying power of the high-power objective (45X) by the magnifying power of low-power objective (10X): then divide this quotient into the diameter of the low-power objective field previously obtained. Do this to find the diameter of the high-power field in microns and millimeters.

9. Repeat steps 7 and 8 for oil immersion objective, if there is one on your microscope. Perform the following steps:

   a. Add a drop of immersion oil to the part of the slide to be observed.

   b. Lower the objective until it touches the drop of oil. You will see a "flash of light" as the drop changes shape.

   c. Focus upward with the fine adjustment head.

10. Complete the computations on the answer sheet.

## Part E. Resolution

Magnification is of importance, but of equal importance is the sharpness of the image. This is the factor called resolution. It is actually defined as the ability to visually separate two closely placed points. This is a function of lens structure called numerical aperture (N.A) and the wavelength (color) of the light being used.

## Part F. Depth of Focus and Plane of Focus

1. Obtain a slide of a tissue, such as mammalian liver, kidney, simple epithelium, or a drop of pond water.

2. Rotate the low power objective into position. Always begin observations with low power before going to high magnifications. Bring the specimen into focus and adjust the light. You will note a number on the fine adjustment head. See how many numerical units are included between being in focus at the bottom of the tissue and at the top of the tissue (bottom and top with respect to the objective lens).

3. Now go to high-dry. Is the whole depth of the tissue cells (or drop of pond, water) which are about 10 microns thick (more or less), in focus at the same time? Is all of the nucleus of a cell in focus at once? Are the bottom and top of the nucleus in focus at the same time? Do the bottom and top of the cell come into focus at the same time? How many numerical units on the fine adjustment head are there between focusing at the bottom of the cell and focusing at the top of it? The plane of focus is usually flat. The depth of focus is approximately the same at the various magnifications and refers to the depth of the plane in focus.

## Part G. Using the Zoom Microscope

If you are to use one of the more simplified microscopes, you will find the instructions for a compound microscope inadequate. On page 9 you will find a diagram of the B&L Academic 225 student zoom microscope. The basic difference between this and the microscope illustrated on page 8 is that there is no nosepiece and therefore only one objective which changes the magnification by the objective between 10X and 50X. The eyepiece is a 10X wide field lens system. The numerical aperture is 0.55.

Although it is built of sturdy, lightweight aluminum construction, it nevertheless must be handled carefully or the lens system will be jarred out of focus. Note the built-in pointer.

Transport the microscope as described in Part B.

Clean the microscope as described in Part C. Surfaces to be cleaned are:

Upper surface of the eyepiece
Objective lens
Upper surface of the light source

### Making an Observation

1. Raise the objective to its highest position. Place a mm ruler or a stage micrometer on the stage over the condenser opening and secure with the clips.

2. Measure the width of the field at the lowest power and again at the highest power.

3. Divide the high power into the low power measurement and multiply by 100. How does this figure compare with the rated magnifications on the instrument?

## Part H. Using the Stereoscopic (Dissecting) Microscope

This is a low power instrument that has a 10X eyepiece (as a rule) and objectives ranging from about 0.7X to 3X magnification. This instrument can be used in both direct and reflected light—that is, the object to be studied is lighted from above. The two eyepieces should be adjusted for the distance between your eyes. When you look through the eyepiece with each eye singly, the image should be in focus (after you have focused with the focusing knob). If the image is in focus for one eye but not the other, turn the eyepiece adjustment band until it comes into focus. When the distance between eyes is properly spaced, the field should appear larger than with either eye singly. Is the image reversed?

Look at a slide of tissue such as one of liver, or pond water.

Can you see cellular details? Examine a penny.

Make a diagrammatic illustration of what you observed. Does a drop of pond water have depth?

## REFERENCES

Gray, Peter, 1967. The Use of the Microscope, McGraw Hill, New York.

Lenhoff, E.S., 1967. Tools of Biology, Current Concepts in Biology Series. The MacMillan Company, New York.

## REPORT SHEET FOR EXERCISE 1.2

### Part A. Observations on the Parts of the Microscope

1. Put the following in the correct order in which light passes on its way from the lamp to the eye by numbering 2, 3, 4, etc.

| | |
|---|---|
| _____ | lamp |
| _____ | nosepiece |
| _____ | eyepiece lenses |
| _____ | Abbe' condenser lens |
| _____ | condenser iris |
| _____ | objective lenses |
| _____ | body tube and draw tube |
| _____ | mirror |

2. Where is the iris diaphragm control located? _____

_____

3. Where is the condenser-raising head located? _____

_____

4. Where is the coarse adjustment head found on your microscope?

_____

### Part B. Making an Observation

5. How does the image move in the field compared with the way that the slide is actually moved on the stage? _____

_____

6. Why must one focus upward on higher powers of magnification?

_____

7. Why do high-dry and oil immersion magnifications require more light than low power? _____

_____

Part C. Resolution

8. Is the bottom of the nucleus in focus when the top of it is? _____

_____

9. Is the top of the cell in focus when the bottom is? _____

_____

10. To what use can the numbers on the fine adjustment head be put?

_____

11. What is the number of your microscope? _____

_____

## RATED MAGNIFICATION

| Objective Lens | Rated Magnification Of Objective | Rated Magnification Of Eyepiece Lenses | Total Rated Magnification |
|---|---|---|---|
| Low Power | _____ | _____ | _____ |
| High-dry | _____ | _____ | _____ |
| Oil Imm. | _____ | _____ | _____ |

Calibration (Real Magnification)

| Objective Lens | Rated Working Distance | Diameter of Field In mm. | microns | Magnification If Lower Power is 100* |
|---|---|---|---|---|
| Low Power | _____ | _____ | _____ | 100X |
| High-dry | _____ | _____ | _____ | _____ |
| Oil | _____ | _____ | _____ | _____ |

*Divide the microns at High-dry and Oil Immersion into the microns across the Low Power Field and multiply by 100. For example:

1,500/1,500 x 100 = 100X

OR,

1,500/375 x 100 = 4 x 100 = 400X

Resolution (Determined from nomogram)

| Objective Lens | Rated N.A. | Color of Light | Resolution in Microns |
|---|---|---|---|
| Low Power | _____ | _____ | _____ |
| High-dry | _____ | _____ | _____ |
| Oil Imm. | _____ | _____ | _____ |

Part D. Using the Zoom Microscope

What is the number of your microscope?

| Rated Magnification of Eyepiece Lens | Indicated Magnification | Implied Magnification of Objective Lenses |
|---|---|---|
| 10X | _____ | _____ (Lowest Power) |
| 10X | _____ | _____ (Highest Power) |

Calibration of the Field (Real Magnification)

| | Diameter of Field In | | Magnification if Lowest Power is 100X |
|---|---|---|---|
| | mm. | microns | |
| Lowest Power | _____ | _____ | 100X |
| Highest Power | _____ | _____ | _____ |

Part E. Using the Stereoscopic (Dissecting) Microscope

1. Is the image reversed?

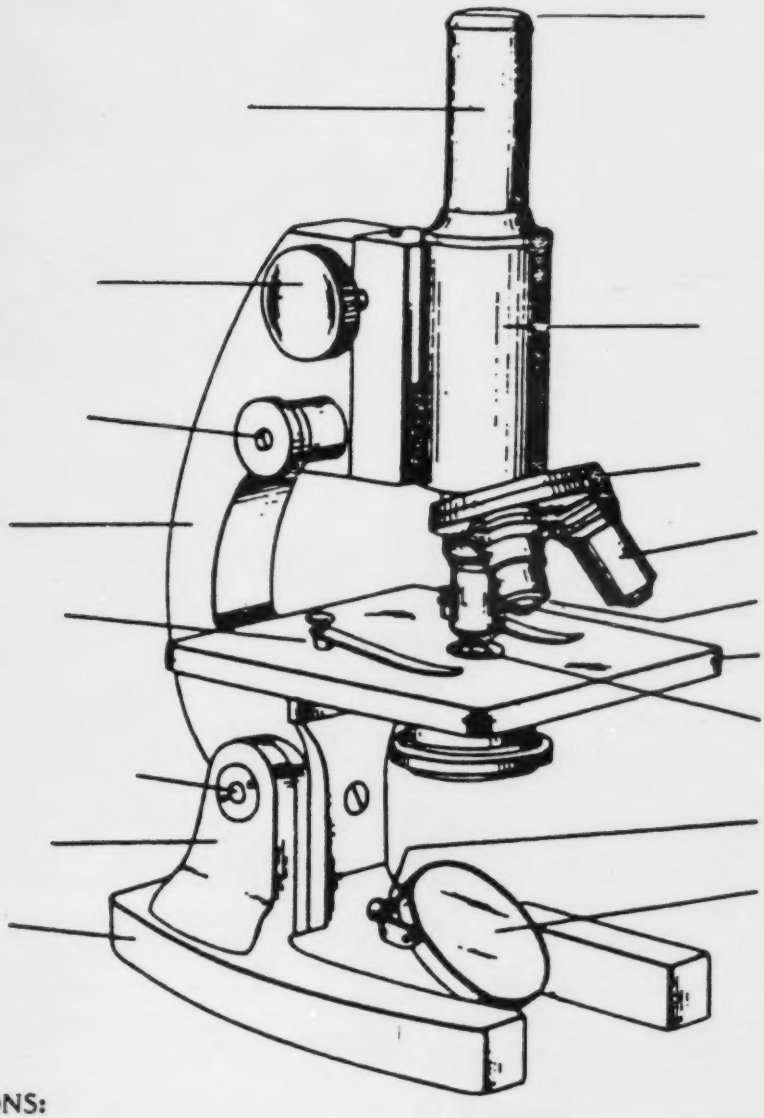
2. How did cells in a slide of tissue look?

3. How did a penny look?

REPORT SHEET FOR EXERCISE 1.2

Self-Test on the Microscope

Can you name the indicated parts of the microscope without referring to the chart on page (  )? Afterwards, check your answers with the diagram and review those parts you missed. Correct any errors.

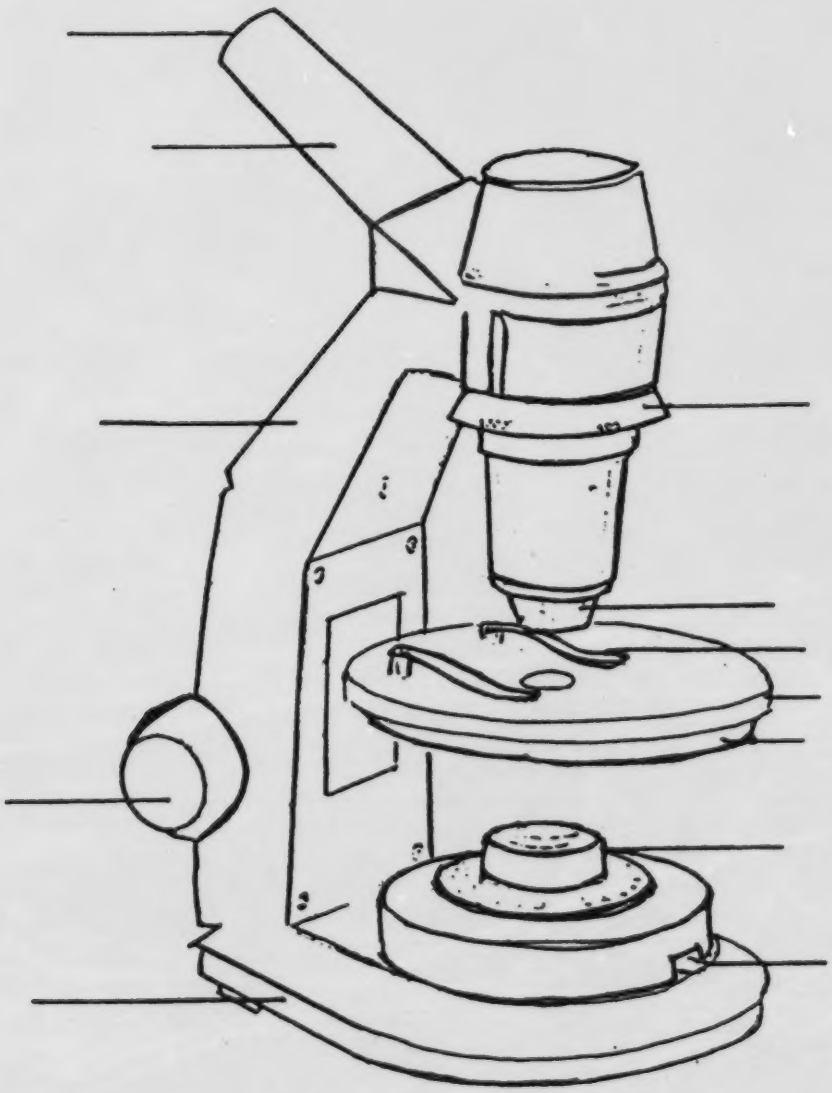


QUESTIONS:

1. Detail some uses of the microscope in the study of biology.

2. What are the limitations of the compound light microscope?

**Self-Test For Zoom Microscope**



Bausch & Lomb
ACADEMIC 255 Student
Zoom Microscope MODEL 011

## EXERCISE 2 - CONCEPTS OF MASS, WEIGHT AND DENSITY

### GENERAL OBJECTIVES:

1. To become familiar with some of the instruments used in making scientific measurements.
2. To become familiar with the metric system.
3. To develop skills for converting British (English) measurements to metric measurements (and vice versa) by using numerical values for each measurement.
4. To develop relevant conceptualizations of mass, weight, density, and volume.

### BEHAVIORAL OBJECTIVES:

Upon completion of this exercise, the student should be able to:

1. Zero a triple beam balance.
2. Weigh specified objects on the triple beam balance.
3. Calculate the volume of a rectangle.
4. Make conversions within the metric system.
5. Determine the density of a solid object.
6. Indicate possible sources of error in measurement.
7. Define the following terms:
   a. Mass
   b. Weight
   c. Density
   d. Volume

8. Measure liquids and compute their relative viscosities.

## The Unit of Distance - Metric System

Every people has had a unit of length. The North American Indians used the pace, the bowshot and a day's journey. When it became necessary to measure off land for irrigation and for plowing, standard rods were made. As early as ancient Egyptian times, when great buildings were made of stone, rather wide use was made of a standard length, a cubit, or the distance from the elbow to the tip of the middle finger. In the times of the Ptolemies, there were professional pacers who helped make maps by pacing out the roads in units called stadia. By medieval times, with the growth of the European nations, there were many measuring units. In England, length was measured by the inch, foot, yard, fathom, rod, furlong, mile, and league. These units reflect convenient early standards.

The French Revolution brought to power a government set sharply against all that was traditional and old-fashioned. An early action of the new government of France was the establishment of a group of learned men ("experts"), to produce a rational set of units for all measurements, the common everyday ones as well as those of science and the blossoming technologies. They set up standards of length, among many others, which have become world-wide in science, and nearly world-wide in everyday life. They called their scheme of units the Metric System, and its fundamental length unit is the meter (from the Greek matron, to measure). They felt it was better to adopt a length standard which had a more lasting significance than the length of the pace, and they were convinced of the value of the decimal system. They therefore chose the meter to be one ten-millionth ($10^7$) of the distance from the equator to the North Pole. In the 1790's, this dimension was rather well-known in terms of carefully laid-out base lines surveyed in Europe. This was the origin of the metric system.

As our standard of length we now employ the standard meter bar made by the French. Many careful copies have been produced. It is worth remembering that a meter is roughly a yard and a foot is about 30cm, while a millimeter is about the thickness of a pencil lead.

## Balance

The simple balance has for a long time given humankind the power to measure quantities of matter even more simply and precisely than space or time could be measured. From Egyptian tombs several thousand years old,

archaeologists have recovered a little balance arm of carved stone with its carefully made stone weights. It was almost surely used then, in the very dawn of history, for the careful measurement of gold dust. The goldsmiths knew even then that the balance was the best way to estimate the amount of solid gold they could cast from any heap of dust or from irregular nuggets.

When an object on one side of a balance is in equilibrium with some standard pieces of matter on the other side, it will remain in equilibrium even though there may be severe changes in the form of the object. Sawing apart a piece of iron, or filing it to a pile of small grains, will not affect the equilibrium of the balance. Let an ice cube melt away to water; the equilibrium remains. A balance responds to some property of matter which is quite independent of the apparent form of the object tested. This property is called gravitational mass. It measures the quantity of matter just as volume measures the amount of three-dimensional space.

Just as we needed to choose a standard meter as the start of our measurement of length, so we need some standard mass to put on one side of the balance with which various other pieces of matter can be compared. The standard used in scientific work is the kilogram. The basic kilogram is a carefully made and guarded cylinder of a durable platinum alloy kept at Sevres, near Paris, along with the standard of length. Here we have a standard which is arbitrary, but reproducible and durable like the standard meter.

Knowledge of the metric system of measurement is most important because within the next few years the United States may be converting completely to the metric system. The U.S. is the only major country in the world which does not already operate on the metric system.

## MATERIALS AND EQUIPMENT

Triple Beam balance
Physiological bars
Aluminum weighing pans
25 or 10 ml graduate cylinders
Metric rulers (cm. and mm.)
Vernier calipers
10 gm masses
Meter sticks
Handbook of Chemistry & Physics

Part A - Weight of an Aluminum Pan and of a Physiological Bar

This experiment will enable you to learn to use the balance and to make conversions within the metric system.

Be sure that the balance is level and that the weights are at ZERO on their scales.

Place the aluminum weighing pan on the balance.

Adjust the weights until the beam swings equidistantly above and below the index mark for the beam end.

Record the weight of the pan (tare weight).

Now add the physiological bar to the weighing pan.

Adjust the weights. Read and record the gross weight.

Return the weights to ZERO and complete the net weight of the bar by subtracting the tare weight from the gross weight.
Gross Weight (pan and physiol. weight) _____ gms.
or _____mgs.

Tare Weight (pan along) _____gms. or
_____mgs.

Net Weight _____gms. or _____mgs.

Mass: Gravitational mass is what we measure on the balance; that is its definition. The balance is to the measurement of mass what the meter stick is to the measurement of length.

Weight: We know that the pull of gravity for any given object is not constant. This pull, which we call weight, varies from place to place on the earth's surface.

Question: Why?

Answer: The gravitational force decreases with distance from the center of the earth. The earth is not a perfect sphere. It is flattened at the equator and at the poles. Also the presence of mountains and valleys on the earth's surface affects gravitational force.

Mass: Is an unchanging property of a body. The weight of a body - the pull of gravity on it - changes very much indeed when the body is moved from the earth to the moon, or to a satellite. Weight and mass are not the same and we must be careful not to confuse the two terms.

Question: If you hold an object in each hand to compare weight, what is it that you are comparing?

Answer: You are comparing the relative pull exerted on the two masses by the earth.

**Part B - Measuring a Solid - Direct Mass Measurement**

Take a mm. ruler and measure the physiological weight for length, depth (or height), and width. Record each measurement.

Length _____mm. Width _____mm. Depth _____mm.

Repeat these measurements using a vernier caliper.

Length _____mm. Width _____mm. Depth _____mm.

Now calculate the volume of the weight using both measurements.

Useful formula: V=length x width x depth.

Question: What advantages or disadvantages do you see in using the two methods of measurement?

Answer: Vernier caliper gives far more accurate measurements because of possibility of getting decimal place figures.

Part C - Density: Indirect Mass Measurement

The ratio of mass to volume is density. It can also be described as weight per unit volume. It is a property of any given kind of matter.

Some objects, like the earth, are too large to put on any balance, yet they possess mass. We can measure the mass in an indirect way. Also, some objects are irregularly shaped and cannot be easily measured. Yet, if we know the mass of the object, we can determine its volume through water displacement.

This experiment is concerned only with finding the volume of a SOLID object.

Purpose: I. To determine the volume of the physiological bar and compare it with the volume calculated in Part B.

DATA:
Original volume + volume with a 10 gm mass
_____ml
Original volume
_____ml
Difference
_____ml
What is the volume of the weight?
What is the density of the weight?
_____g/ml

(You find the density by dividing the weight of the 10 gram mass by its volume.)

Of what metal is the 10 gram weight composed?_____
_____

For this question refer to the table below.

Question: What possible sources of error exist in the experiment?

1. scales not accurate
2. measurements not accurate
3. splashed water out of cylinder while doing volume measurements.

## EXCERPTS FROM THE
## DENSITY OF ELEMENTS & DENSITY OF ALLOYS TABLE*

| NAME | DENSITY (SPECIFIC GRAVITY) | THERMAL EXPANSION COEFFICIENT PER $^{o}C$ | MELTING POINT $^{o}C$ |
|---|---|---|---|
| Aluminum (2S) | 2.699 | $23.94 \times 10^{-6}$ | 660 |
| Aluminum bronze (copper & aluminum) | 7.69 | 16.5 | 1050 |
| Brass, Yellow (copper & zinc) | | 8.44 | 940 |
| Lead (white metal) | 9.43 | | 238 |

*Above figures were taken from the "Density of Elements" (p.2185) and "Density of Alloys" table (p.2187) in the Handbook of Chemistry and Physics, 44th ed., 1962.

II. To determine the volume of an irregular object:

METHOD: (1) Add 5 milliliters (ml.)* of water to a graduate cylinder. Be sure of accuracy. Read the base of meniscus. (The meniscus is the curved upper surface of a liquid column.)

Place the physiological bar in the graduate cylinder by letting it slide gently down the side of the cylinder being careful not to splash the water.

Read the volume at the base of the meniscus.

Record it.
*ml = cc (cubic centimeter)

Original volume+volume with weight _____ml

Original volume (without weight) _____ml

DIFFERENCE _____ml

The difference equals the volume of the bar. How does this figure compare with the figure for volume that you calculated for Part B?

_____

What is the density of the physiological bar?_____
_____gms/ml.

(2) Add 5 ml of water to a graduate cylinder.

Place the 10 gram mass in the cylinder by letting it slide gently down the side, again being sure not to splash water out of the cylinder.

Read the final volume. Record it.

(3) Fill the pipette exactly to the 1.0 ml. mark with water.

Determine the time in seconds that it takes for the water to run out. Repeat this four more times. Average your results.

Using the same pipette, repeat the above procedure using alcohol, then do it with glycerol, then soft drinks.

Before starting the trials with a new solution, rinse out the pipette with some of the next solution, so that you will measure the behavior of a homogeneous liquid. Repeatability depends upon the accuracy with which you fill the pipette. Using the time that it took water to run out of the pipette as a standard, (for example, 1.000), compute what the relative viscosities of alcohol and glycerol would be by dividing the time required for the water to run out into the times for the other fluids.

| Trial | Water | Alcohol | Glycerol |
|---|---|---|---|
| 1 | _____ | _____ | _____ |
| 2 | _____ | _____ | _____ |
| 3 | _____ | _____ | _____ |
| 4 | _____ | _____ | _____ |
| 5 | _____ | _____ | _____ |

Sum
Average or Mean

Relative Viscosity:

Water 1,000
Alcohol _____
Glycerol _____

Questions:

1. What is the difference between scales and a balance?

2. Is gravity the same all over the earth?

3. Why are balances used in scientific laboratories for weight determinations instead of scales?

4. How many grams are in an ounce? _____

5. How many grams are in a pound? _____

6. How many grams are in a kilogram? _____

7. How many pounds in a kilogram? _____

8. How many centimeters in an inch? _____

9. How many centimeters in a meter? _____

10. A test tube may be 14 mm. x 140 mm. These measurements converted to inches will be: _____in. x _____in.

11. What particles of the atom account for most of its weight?

12. Distinguish between the mass of an object and its weight.

13. Relate your body to a tree. Which has the greatest:

Mass? Weight? Density?

Indicate the basis for your statement in each instance.

REFERENCES

PHYSICS - Physical Science Study Committee (2nd Edition) D.C. Health & Co., 1965.

Handbook of Chemistry & Physics, 44th ed., 1962

## APPENDIX A

| Prefix | Meaning | Numerical Value | Symbol |
|---|---|---|---|
| Mega- | One Million | $10^6$ or 1,000,000 | M |
| Kilo- | One Thousand | $10^3$ or 1,000 | K |
| Deci- | One Tenth | $10^{-1}$ or 0.10 | d |
| Centi- | One Hundredth | $10^{-2}$ or 0.01 | c |
| Milli- | One Thousandth | $10^{-3}$ or 0.001 | m |
| Micro- | One Millionth | $10^{-6}$ or 0.000001 | μ |
| Nano- | One Billionth | $10^{-9}$ or 0.000000001 | n |

Therefore, multiples or fractions of a gram would be indicated by adding the appropriate prefix to the gram, viz: kilogram, milligram, or microgram.

| Unit of Weight | Comparative Weight | Abbreviation of Symbol |
|---|---|---|
| Kilogram | $10^3$ gm | Kgm |
| Gram | 1 gm | Gm. |
| Milligram | $10^{-3}$gm | Mg. or mgm. (or gamma) |
| Microgram | $10^{-6}$gm | gγ |

| Unit of Length | Comparative Length | Abbreviation of Symbol |
|---|---|---|
| Kilometer | $10^3$ m | Km |
| Meter | 1 m | M |
| Centimeter | $10^{-2}$ m | cm. |
| Millimeter | $10^{-3}$ m | mm. |
| Micron | $10^{-6}$ m | μ |
| Millicron (Nanometer) | $10^{-9}$ m | m (n) |
| Angstrom unit | $10^{-10}$ m | $\overset{o}{A}$ |

SAMPLE CONVERSIONS

(a) To convert 25 cm to meters

If 100 cm = 1 m

25 cm = x meters

$\frac{100}{1} = \frac{25}{x}$

$100x = 25$

$x = 0.25m$

So, 25 cm = 0.25 m

(b) To convert 25 cm to millimeters

If 1 cm = 10 mm

25 cm = x mm

$\frac{1}{10} = \frac{25}{x}$

$x = 250$ mm

So, 25 cm = 250 mm

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## APPENDIX B

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Useful Formulae

Area of a:

Square or rectangle = length x width

Triangle = 1/2 base x height

Trapezoid = base x altitude

Circle = $\pi r^2 = 1/4\pi d^2$ ($\pi = 3.1416$)

Volume of a:

Cube or rectangular block = length x width x height

Prism = length x width x altitude

Cylinder = $\pi r^2$ x altitude

Cone or pyramid = $1/3\pi r^2$ x altitude

Sphere = $4\pi r^{2/3}$

## EXERCISE 3.1 - TOOLS FOR THE STUDY OF BIOLOGY

### GENERAL OBJECTIVES:

1. To familiarize students with some of the tools used in the science of biology.
2. To develop skills in the use of tools for studying biology.
3. To enable students to use the metric system.
4. To develop skills in measuring and using numbers.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students will be able to:

1. Write the names of the tools on the table used for studying biology.
2. Write out the uses of these tools.
3. Carry out manipulative functions of laboratory instruments for observation and measurements found on the table, in the presence of the instructor.
4. Demonstrate skills in the usage of units of measurement in the metric system by measuring specific objects on the table with the metric ruler, the pipette, the balance tools and the graduated cylinder in the presence of the instructor.

### INTRODUCTION

Biology today requires a basic knowledge of the use of a variety of tools and instruments. Use of the microscope alone is no longer sufficient in preparing students for the types of activities encountered in a modern biology class laboratory. The ability to weigh materials and measure length and volume accurately is an important skill necessary for the successful study of modern biology. This exercise will introduce the student to some of the tools of modern Biology and will develop the necessary skills for measurement.

## MATERIALS AND EQUIPMENT

- Dissecting kits
- Petri dishes
- Microscopes
- Balances
- Scales
- Thermometers
- Beakers
- Graduated cylinders
- Ehrlenmeyer flasks
- Test tubes
- Test tube racks
- Test tube clamp
- Watch glass
- Spatula
- Graph paper
- Metric rulers
- Slides and cover-slips
- Medicine droppers
- Pipettes
- Funnels
- Stirring rods
- Culture dishes
- Wash bottles
- Bunsen burners
- Ring stand
- Mortar and pestle
- Spot plates
- Transfer loops
- Brushes

If available, add:

autoclave, drying ovens, microtome, centrifuge, spectrophotometer, pH meter, kymograph, burettes, seine nets and ecological collecting equipment, and other available laboratory equipment.

Objects of various sizes and weights.

## PROCEDURE

### Part A

On the table we have a variety of tools used in the study of biology, some simple and some rather sophisticated. Select some tools and look at their forms and shapes. Compare these to other tools or objects you have seen in other places and guess how they could be used.

### Part B

You have available a variety of tools and instruments on the table. Use whatever tools you need to solve the following problems:

1. Determine which is the heaviest of several objects of approximately the same weight but which vary in size.

2. Determine the length and width of several of the objects provided and express these dimensions in metric units.

3. How would you clean bottles, test tubes, and pipettes?

4. How would you determine the concentration of colored solutions?

5. Take equal amounts of solution provided and place them into five test tubes.

6. Determine the temperature of the water in the beaker.

## UNITS OF SCIENTIFIC MEASUREMENT

### Prefixes

The common unit of measurement for weight in the scientific laboratory is the gram. This unit may be subdivided into smaller units for greater accuracy in weighing small objects, or be multiplied to give larger units for convenience in weighing heavy objects. Prefixes are used to designate multiples or fractions of a standard weight, distance, or other quantity.

| Prefix | Meaning | Numerical Value | Symbol |
|---|---|---|---|
| Kilo | One thousand | $10^{3}$ or 1,000 | K |
| Deci | On tenth | $10^{-1}$ or 0.1 | d |
| Centi | One hundredth | $10^{-2}$ or 0.01 | c |
| Milli | One thousandth | $10^{-3}$ or 0.001 | m |

| Units of Weight | Comparative Weight | Abbreviation or Symbol |
|---|---|---|
| Kilogram | 1000 gm | kgm |
| Gram | 1 gm | gm |
| Milligram | .001 gm | mg |

| Units of Length | Comparative Weight | Abbreviation or Symbol |
|---|---|---|
| Kilometer | 1000M | km |
| Meter | 1 M | m |
| Centimeter | .01 M | cm |
| Millimeter | .1 cm or .001 M | mm |
| Micron | .001 mm or .000001 M | μ or mμ |

| Units of Volume | Comparative Weight | Abbreviation or Symbol |
|---|---|---|
| Liter | 1 L | L |
| Milliliter | .001 L | ml |
| Cubic Centimeter (cc) | 1 cm x 1 cm x 1 cm | $cm^3$ |

| Units of Temperature | Degrees Celsius Formerly Centigrade | Farenheit° Equivalent |
|---|---|---|
| Temp. of boiling water | 100° | 212° |
| Body Temperature | 37° | 98.6° |
| Room temperature | 23° | 73° |
| Temp. of water freezing | 0° | 32° |

Formulae

Area of a square or rectangle = length x width
Volume of a square or rectangle = length x width x height
Density = weight/volume

QUESTIONS

1. List 10 of the most important tools used in biology and indicate how they are used.

2. Which are more important in a biological laboratory, tools or measurements?

3. Explain the advantages of the metric system over the English system of measurement.

## REFERENCES

Lenkoff, E.S., Tools of Biology, Current Concepts in Biology Series. The MacMillan Company, New York.

## EXERCISE 3.2 - CELL TYPES

### GENERAL OBJECTIVE:

To provide specific examples of cells in order that the student might formulate generalizations about the structure and organization of cells.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students should be able to:

1. Prepare a wet mount of a given organism for observation with the microscope.

2. Given a diagram of the cell, identify and name the main structural parts of a cell 3.Write a paragraph comparing and contrasting the cell types observed.

4. Demonstrate, through the labeling of drawings, the organization of cells into tissues and/or organs into multicellular plants and animals.

### INTRODUCTION

It has been a frequent custom in the past to draw a diagram of a cell which includes as many features as desired from the study of a variety of cells at different magnifications, and then label this a "typical cell". Of course, such a cell diagram would be hardly typical. There are, instead, a number of cell varieties, each one typical of the variation of that type. All cells, however, seem to hold a number of structures in common. We shall look at several types of cells in this exercise. The student should study each one and then reach some generalizations (conclusions) about what is typical of living and preserved cells.

## MATERIALS AND EQUIPMENT

Microscope and lamp
Microscope slides and
coverglasses (coverslips)
Petrolatum (Vaseline) gun (a 2cc
syringe without needle)
Syracuse watch glass
Medicine dropper
Neutral Red or Methylene Blue
solutions
Forceps, scissors, dissecting needle
and single-edged razor blade
5% methyl cellulose

Living Cultures: Fresh onions, Elodea sprigs, Nitella, baker's yeast, Amoeba

Prepared slides: Mammalian liver stained with haematoxylin and eosin, mitochondria in liver cells, leaf types, bull sperm, Ascaris megalcephala sperm entrance.

### Slide Preparation

Two precautions in making temporary mounts: The coverglass must be supported a slight distance above the specimen so as not to mash it. The specimen must be kept wet. This may be done by adding more fluid to the edge of the coverglass or by sealing the fluid under the coverglass by using a petrolatum barrier to prevent evaporation.

### Simple Wet Mount

Using the petrolatum gun, lay down a circle of petrolatum about the size of a dime . The opening of the circle permits excess fluid to escape as the coverglass is pushed close to the slide. The specimen is transferred to the circle in a small drop of fluid and covered with a coverslip.

### Open-Ended Wet Mount

Lay down two parallel lines of petrolatum the length of the coverglass. Transfer the specimen to the enclosed space with a dropper. Cover it with the coverslip. Fluids beneath the coverglass can then be exchanged by applying a drop to one side and removing the excess fluid with paper towel (or bibulous paper) from the opposite side. Fluids must be replaced often to prevent the specimen from drying out. Be careful not to lose specimen when exchanging fluid.

## Sketches and Drawings

1. Place sketches and drawings below the legends provided.

2. Place drawings to the left and label neatly to the right side.

3. Make drawings and sketches large enough to show details easily. Most drawings should be at least 1½ inches across.

## Plant Cells

1. Onion Epidermal Cells. Remove a fresh scale from an onion. Remove the inner epidermal layer with a razor blade and forceps and transfer it into the water in a Syracuse watch glass. Cut it into suitable pieces with sharp scissors and transfer them to an open-ended wet mount. Observe them unstained with low illumination. You should be able to see the cell wall, central vacuole and perhaps the thin layer of cytoplasm adjacent to the cell wall. Now draw some methylene blue stain under the coverslip, followed by several drops of tap water to wash away the excess stain. You should now be able to see the nucleus. Sketch one cell and label the cell wall, cytoplasm, nucleus and vacuole.

2. Cytoplasmic Streaming in Nitella. Make a simple wet mount of a few filaments of the alga Nitella. Observe under high-dry magnification with dim illumination. Is there any movement to be seen in these cells? Diagram a cell, labeling its parts, and indicate the direction of any movement observed in the cytoplasm.

3. Cytoplasmic Streaming in Elodea. Choose a young leaf near the end of a spring of Elodea (the water weed). Remove it with forceps, being careful to leave the top of the leaf unbruised. Make a simple wet mount and observe under the microscope with bright illumination. Sketch two or three continuous cells and indicate in which directions movement is observed. Label the vacuole, cytoplasm, chloroplasts and the cell wall.

4. Palisade Cells From A Green Leaf. In your slide collection find the slide labelled "Leaf Types". Study and compare the structure of these three types of leaves. Sketch a palisade cell from a dicot leaf. Label the nucleus, chloroplasts, cytoplasm and vacuole.

Microbial Cells

5. Yeast Cells. To a drop of active yeast culture on a microscope slide add a drop of methylene blue and add a coverslip. Sketch a cell and label the features observed.

Animal Cells

6. Amoeba. These organisms appear as white specks on the bottom of the culture dish. Use a medicine dropper to aspirate one or more and transfer them to a Syracuse watch glass. Prepare a simple wet mount and add Neutral Red stain under the coverslip. Note the way in which the amoeba moves.

   Identify the parts of the cell. How can the movement of the amoeba be explained?

   Make a sketch of the amoeba, indicating the following: the plasmalemma, the plasmagel, the plasma sol, pseudopods. Indicate with arrows the direction of the movement of the cytoplasm.

Set the slide aside and look at it again in about twenty (20) minutes. Where is the Neutral Red dye located in the cell?

   How can this be explained?

7. An Animal Egg Cell. Find a slide of Ascaris eggs. These are probably the most idealized of the cells we will look at today. They are protected by a gelatinous coat. The cell limit is visible. Draw an egg. Label all of its parts.

8. A Sperm Cell. Find the slide labelled "Bull Sperm". These are rather typical mammalian sperm cells. Study them under oil immersion and identify the acrosome derived from Golgi material, the head (nuclear material), the ring centriole, the midpiece which contains many mitochondria, and the tail filament which is a long flagellum arising from a centriole. Did you see a centriole in the egg cell? Look again. Find the slide labelled "Ascaris megalocephala, sperm entrance". Can you identify the sperm cell of Ascaris? How is it different from bull sperm? Does it have a centriole? Do ascaris eggs have a centriole? Find an example where the sperm has entered an egg. Does such an egg have centrioles?

9. Mammalian Liver. There are many similar cells in a tissue like this. Make a large drawing of a single cell. Label the cell limit, the cytoplasm, the nucleus, and any other features you can identify.

10. Mitochondria. Make a drawing of similar size to that in 9, indicating the locale of the mithochondria and their shape.

## REFERENCES


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Kimball, John W. 1970. Cell Biology, Addison-Wesley, Readi ng Mass.

Readings from Scientific American, 1965. The Living Cell, Freeman, San Francisco.

Simpson, G.S. and W.S. Beck. 1969. Life, short ed. Harcourt, Brace and World, New York.

## REPORT SHEET FOR EXERCISE 3.2

1. Onion epidermal cells taken from the inner surface of an onion scale and stained with _____.
   Drawn from _____X.

2. Nitella Cell. The arrows indicate the direction of movement observed.
   Drawn from _____X.

3. Elodea Leaf Cells. Arrows indicate the direction of movement observed. Drawn from _____X.

4. Palisade cell from the mesophyll layer of a green leaf.
Drawn from _____X.

5. Yeast cells stained with methylene blue.
Drawn from _____X.

6. Amoeba cells supravitally stained with Neutral Red. Arrows indicate the direction of movements seen in the cell. Drawn from _____
_____X.

7. Amoeba _____ minutes after being stained with Neutral Red.
Drawn from _____X.

8. Egg cell of _____.
Drawn from _____X.

9. Bull spermatozoan cell. Drawn from _____X.

10. Cells from the mammalian liver. Fixed and stained with haematoxylin and eosin. Drawn from _____X.

11. Mitochondria demonstrated in the liver of the _____
Drawn from _____X.

QUESTIONS

1. State the Cell Theory.

2. What are the implications of the Cell Theory?

3. What is our course description of a cell? Do all cell studies today conform to this description?

4. Bull spermatozoa do not divide and they have an unusual shape. What arguments can be made for calling them cells within our course definition?

5. Were you able to see a centriole in an unfertilized Ascaris egg? In a fertilized egg? Where did it come from?

6. How can one explain the streaming of the cytoplasm observed in Nitella or Elodea?

## EXERCISE 4 - DIFFUSION, OSMOSIS AND ACTIVE EXCLUSION

### GENERAL OBJECTIVES:

1. To enable students to understand the physical principles involved in simple diffusion, osmosis, plasmolysis and active exclusion.

2. To assist students in relating concepts about the movement of materials to the activities of cells.

3. To provide students with an opportunity to graph the physical phenomena of diffusion, thereby increasing their understanding of space-time relationships.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students should be able to:

1. Explain the concept of diffusion, utilizing the information recorded on the report sheet relative to this topic.

2. Explain how space-time relationships portrayed by graph curves represent the osmotic process.

3. Make out a table explaining the differences between simple diffusion, osmosis and active exclusion.

4. Draw a diagram comparing a normal cell with a plasmolyzed cell.

### INTRODUCTION

Substances enter the cell by way of the cell membrane, and after entering they move from one site to another within the cell. Waste products and secretions that result from cellular activities also exit by way of the cell membrane. This exercise is designed to facilitate an understanding of the manner by which these substances move into and out of the cell.

## MATERIALS AND EQUIPMENT

1/4 inch cellophane dialysis tubing
10% gelatin
bromthymol blue (0.016%)
0.1 N HCl
0.1 N NaOh
test tubes
white potatoes
graduate cylinders
towels
elodea leaf
NaCl solution (10%)
Lugol's iodine solution
normal saline solution (0.9%)
0.1 M iodoacetate solution
0.1 M KCN Solution
0.5% Congo Red
filter paper
bunsen burners
yeast cell preparation

## PROCEDURE

### Part A. Diffusion of Hydrogen and Hydroxyl Ions Through Gelatin

1. Cut a 12-inch length of 1/4-inch cellophane dialysis tubing and soak it in a beaker of water for a few minutes until it becomes soft. Work open the ends, insert a small funnel, and fill with tap water. Hold the ends and squeeze the water toward the center of the loop. If there are any holes in the tubing, a fine stream of water will be expelled. Such a piece of tubing must be replaced.

2. To a hole-free piece of tubing add about 6 inches of melted 10% gelatin containing enough bromthymol blue (0.16%) to give it a definite color. Center the gelatin in the tubing and insert into a beaker containing 0.1N HCl. Note the time you put the tube into the acid and the time it takes to turn all to one color. What is the color?

3. Remove the bag of gelatin from the acid. Rinse it off in tap water and transfer it to a beaker of 0.1N NaOH. Note the time you put it in NaOH and the approximate time for it to change color. What would a green band indicate? What is the final color?

4. Answer the questions for the section on the Report Sheet below.

## Part B. Osmosis

1. Obtain 7 test tubes and mark them 0%, 1%, 2%, 3%, 4%, 5%, and 10%. Fill them about 3/4 full of the corresponding concentration of NaCl solution.

2. Cut seven plugs of white potato with a cork borer. Size 1-3 is suggested. The plug must fit easily into the 10 ml. graduated cylinders you will use to measure the volume of each plug.

3. Measure the volume (using tap water) and weigh each plug, then transfer it to a tube of NaCl. Record the weight, volume, tube number and starting time on your Report Sheet.

4. After 90 to 120 minutes, recover the potato plugs from each tube (one at a time so they do not get mixed up). Blot away the excess solution with a damp (not wet) piece of paper towelling and determine its weight and volume again. How does each plug feel? Record your data. Return the plugs from 0% and 10% to their tubes of NaCl for use in the next section of this exercise. Discard the others.

5. Plot the data for volume and weight on the graphs provided (Graphs 1 and 2).

6. Plot the data for percent change in weight against the reciprocal of the NaCl concentration (Graph 3). Do the same for the percent change in volume (Graph 4).

   What accounts for the shapes of the curves observed in Graphs 1 and 2?

## Part C. Plasmolysis in Elodea Leaf

1. Make an open-ended wet mount of an Elodea leaf so that fluids can be exchanged under the coverglass (see Exercise No. 5).

2. Find a green cell and sketch it.

3. Draw a 10% NaCl solution under the coverglass, drawing water off the other side with a bit of paper towelling.

4. Sketch the same cell at 1 minute intervals for 5 minutes. What happens to the vacuole? The plasma membrane? The cell wall?

5. Now add water to one side of the slide and draw off the salt solution. Wash the leaf well in this way. Now sketch the cell at 1 minute intervals for 5 minutes.

6. Take a single-edged razor blade and cut a thin wedge from the potato plug in water (0% NaCl). Add a drop of Lugol's iodine solution to it in a Syracuse watch glass. Drain and rinse with water. Make a wet mount and observe under the microscope. The iodine will have stained the starch grains dark blue or black. Identify the cell wall, vacuole and the starch grains. Sketch. Repeat this procedure with the potato plug in 10% NaCl, remembering to use 10% NaCl to rinse away the Lugol's solution. Sketch a cell and label as above.

7. Answer the questions for this section on the Report Sheet below.

Part D. Active Exclusion of a Substance

1. Label 3 test tubes: N (normal), B (boiled), and P (poisoned).

2. Add about 5 ml of a growing yeast suspension to each test tube.

3. Place tube B in a container of boiling water or heat over a bunsen burner until the tube contents boil for at least 15 seconds. This will kill the cells.

4. Add 5 drops of 0.01 M KCN and 5 drops of 1 M iodoacetate to the tube labeled P. (REMEMBER: Both KCN and iodoacetate are poison. If you get any on your hands, wash them immediately).

5. Now, to each tube add enough 0.5% Congo Red to give a good color. Record the color in each tube on your report sheet.

6. Examine a drop of each suspension under the microscope, starting with tube N. What colors are the cells in each tube? Are all of those in tube N stained? How about tubes B and P?

7. Place a small funnel into each of three clean test tubes. Prepare a filter paper for each. Filter the remaining contents of each tube. Record the color of the cell on the filter paper and the filtrate that comes through.

Turn in your Report before you leave for the day.

## REFERENCES


Holter, H., 1961. "How Things Get Into Cells", Scientific American, Reprint No. 96, (September).

Solomon, A.K., 1960. "Pores in the Cell Membrane", Scientific American, Reprint No. 76.

Wolf, A.V., 1968. "Body Water", Scientific American (November).

## REPORT SHEET FOR EXERCISE 4

Part A

Starting color:_____ Color in 0.1 N HCl:_____

Time into the acid_____ Time color complete_____ Minutes_____

Radius of the tubing is_____ inches or_____ cm. or_____ mm

What is the approximate diffusion rate?____ cm./sec. or_____ cm./min.

Color in alkali_____.

Time into 0.1 NaOh_____ Time color complete_____ Minutes_____

Approximate diffusion rate is_____ mm./sec. or_____ cm./min.

Did you observe a green band? What did it indicate?

What caused the hydrogen ions to move into the gelatin?

According to kinetic theory, in what directions are the ions moving?

What is meant by the flux of an ion?

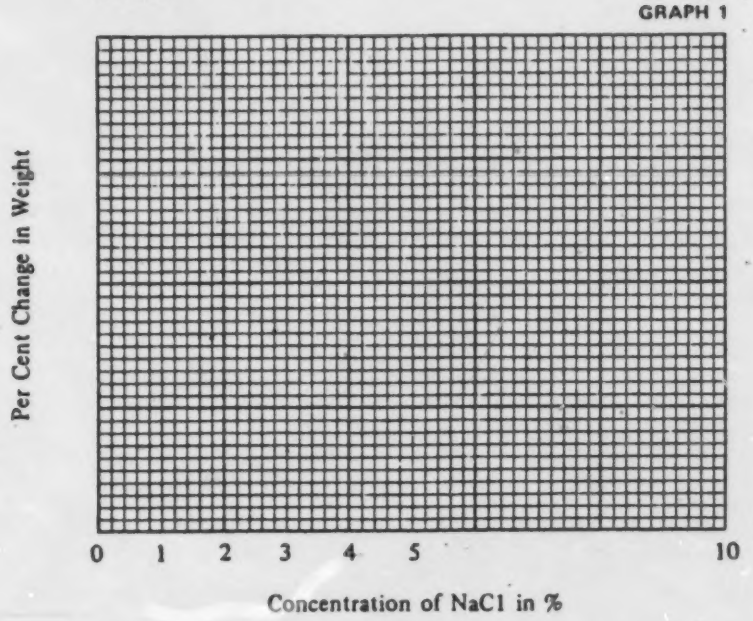
In what ways does a cellophane (cellulose acetate) membrane differ in its properties from a plasma membrane?

| | TIME EXPOSED TO NaCl:_____ MINUTES | | | | | | |
|---|---|---|---|---|---|---|---|
| Conc. of NaCl: | 0% | 1% | 2% | 3% | 4% | 5% | 10% |
| Starting weight of potato plug in mg. | | | | | | | |
| Final weight of potato plug in mg. | | | | | | | |
| Change in wt. (mg.) | | | | | | | |
| Starting volume of potato plug in cc. | | | | | | | |
| Final volume of potato plug in cc. | | | | | | | |
| Change in vol. (cc.) | | | | | | | |
| % Change in volume | | | | | | | |
| How does the plug feel at the end of the experiment? | | | | | | | |

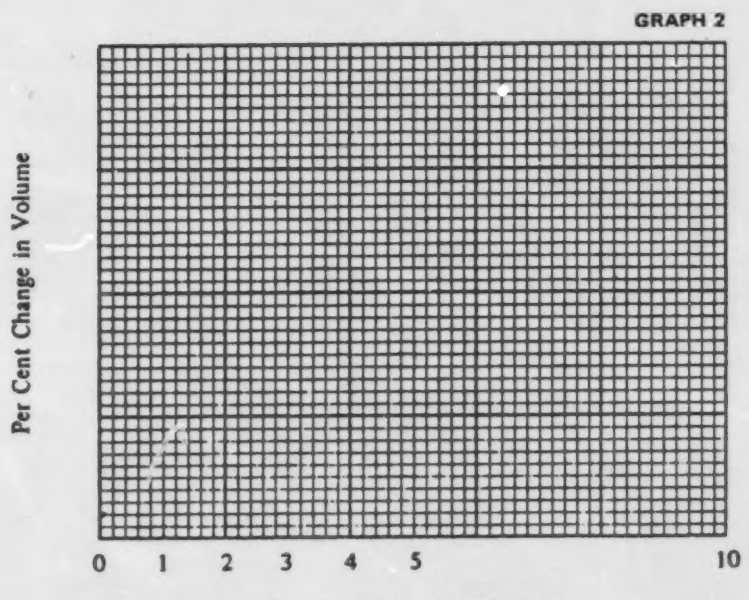
Part A Report Sheet Contd.

Osmosis

GRAPH 1



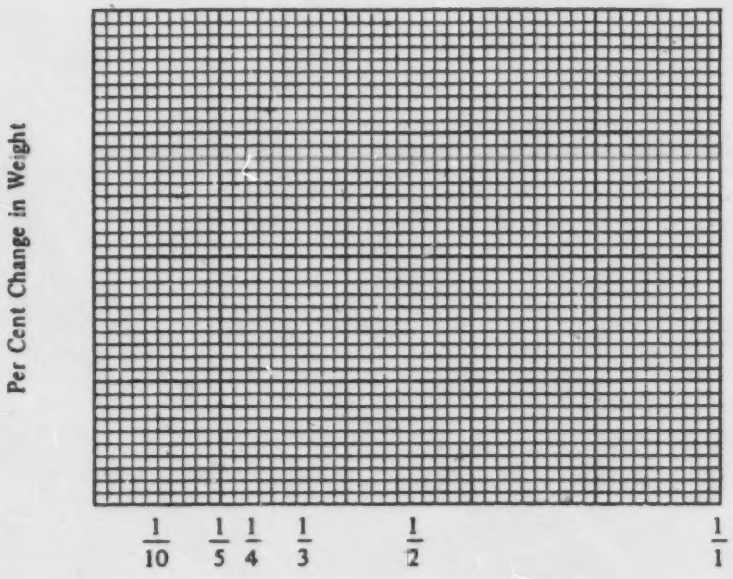

GRAPH 2



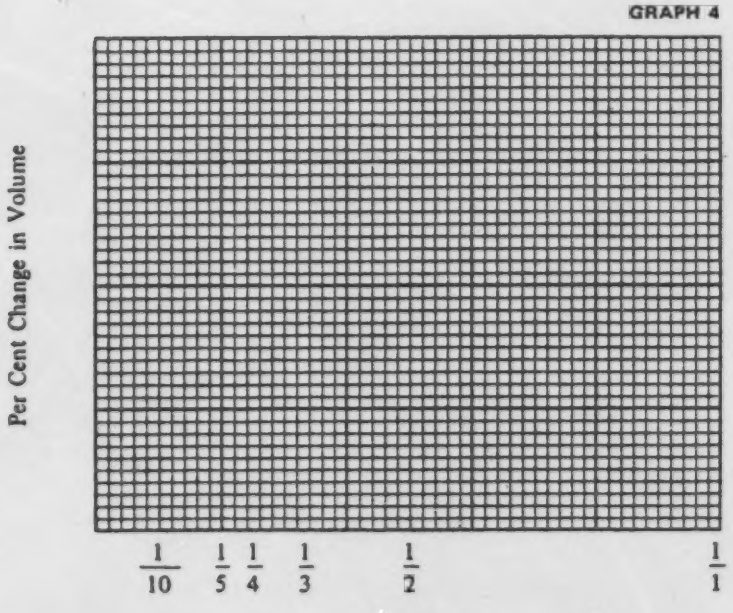
Part A Report Sheet Contd.

Osmosis

GRAPH 3




GRAPH 4



## QUESTIONS

1. Plasmolysis literally means "to dissolve the cytoplasm". Did this occur?

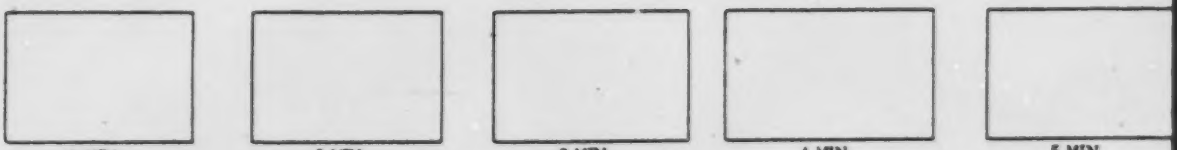
2. How did the cell wall respond to NaCl?

3. What response or what behavior was exhibited by the plasma membrane (cell limit)?

4. Why did the vacuolar contents behave as they did?

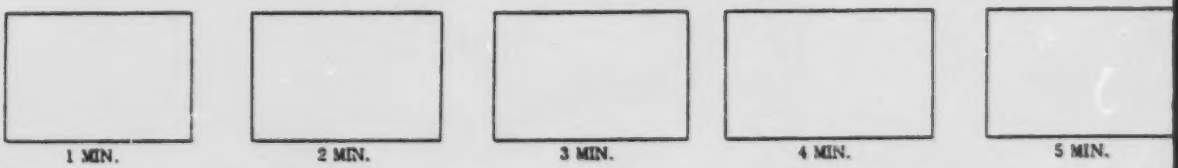
### Plasmolysis in the Elodea Leaf

START



1 MIN. 2 MIN. 3 MIN. 4 MIN. 5 MIN.

Time after adding 10% NaCl.



1 MIN. 2 MIN. 3 MIN. 4 MIN. 5 MIN.

Time after washing with distilled water.

Sketch of cell from potato plug kept in 0% NaCl and stained with Lugol's iodine solution.

Sketch of cell from potato plug kept in 10% NaCl and stained with Lugol's iodine solution.

## REPORT SHEET FOR EXERCISE 4

### REPORT SHEET

| | TUBE N | TUBE B | TUBE P |
|---|---|---|---|
| Percent of cells in the field that are stained | | | |
| Color of cells stained under the microscope | | | |
| Color of the tube contents | | | |
| Color of the filtrate | | | |
| Color of cells on the filter paper | | | |

What color changes did you find with change in pH for Congo Red in Experiment 3?

How do you explain the number of cells stained in Tube N?

How do you explain the number of cells stained in Tube B?

How do you explain the number of cells stained in Tube P?

Would you conclude from your observations that Congo Red can pass through the cell membranes of yeast? Why?

Did the cell membrane protect the cell from a harmful substance (KCN)?

What is the difference in the mechanisms operating in tubes B and P?

QUESTIONS

1. Why do all living cells have membranes which are permeable to some substances?

2. What is the difference between the manner in which substances move across a living membrane as opposed to a non-living membrane?

3. What are some factors which influence the rate of diffusion?

4. How would you describe the structure of the cell membrane?

5. Why is it necessary to rinse a blood syringe with a physiological saline solution as opposed to a hypotonic or hypertonic solution?

## EXERCISE 5 - WATER CONTENT OF VARIOUS TISSUES AND CELLS

### GENERAL OBJECTIVE:

1. To demonstrate the importance of water as a constituent of protoplasm.

2. To develop communication skills.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students should be able to:

1. Determine the water content of a given tissue.

2. Make a chart showing the comparison of the amounts of water found in different types of plant and animal tissues.

3. Exhibit the skill of communicating by constructing and explaining an appropriate bar graph.

### INTRODUCTION

Water is necessary for the maintenance of life. Many living organisms cannot tolerate more than a 25-33% water loss. Water is important because many substances dissolve in it. It holds and conducts heat well and its solutions of salt are good conductors of electricity.

In this exercise, the class will investigate and compare the amounts of water found in plant and animal tissue.

### MATERIALS AND EQUIPMENT

Drying oven
Knives or scalpels, forceps
Triple beam balance
3 aluminum weighing plans per student
Food items (as described below)

30-35 grams of these suggested food items: potatoes, carrots, onions, spinach leaves, dry beans, nuts, yeast cake, liver, kidney, fat and bone.

## PROCEDURE

1. Mark each pan with a number and weigh as accurately as you can. Record the weight as tare weight.

2. Place 10-20 grams of the material assigned to you (cut into very small pieces) in the pans and reweigh. Record as gross weight.

3. Subtract the tare weight from the gross weight to obtain the net wet weight for each sample.

4. Place pans containing samples in the drying oven set at 100-110°C to dry overnight or until all your samples have dried to a constant weight. Record this as gross dry weight.

5. Subtract the tare weights and determine the net dry weights.

6. To determine how much water was lost from the tissue, subtract the net dry weight from the net wet weight. Divide the net wet weight of each tissue into the weight of the water lost and multiply by 100. This will give the percent of water in each tissue.

e.g.:

Net Wet Weight = 20 gm

Net Wet Weight = 15 gm

Water loss = 5 gm

Percent Water in Tissues =

$$\text{(water loss/net wet weight)} \times 100 = \frac{5}{20} \times 100 = 25\%$$

## REPORT SHEET FOR EXERCISE 5

The Water Content of Various Tissues

Material Used:

| | Pan 1 | Pan 2 | Pan 3 |
|---|---|---|---|
| Gross Wet Weight | _____ | _____ | _____ |
| Tare Weight | _____ | _____ | _____ |
| Net Wet Weight | _____ | _____ | _____ |
| Gross Dry Weight | _____ | _____ | _____ |
| Tare Weight | _____ | _____ | _____ |
| Net Dry Weight | _____ | _____ | _____ |
| % Water | _____ | _____ | _____ |

Computation of the Mean (Average)

| Pan # | % Water |
|---|---|
| 1 | _____ |
| 2 | _____ |
| 3 | _____ |
| Sum | _____ |
| Mean (sum/3) | _____ |

(Optional) Computation of the Standard Error of the Mean

| Pan # | Deviation (d) from Mean | $d^2$ |
|---|---|---|
| 1 | _____ | _____ |
| 2 | _____ | _____ |
| 3 | _____ | _____ |
| Sum | _____ | _____ |

Variance = _____

Standard Deviation = _____

Standard Error of the Mean = _____

Compare class data for all materials tested and rank materials according to water content.

| | Material | %Water Content |
|---|---|---|
| 1. | _____ | _____ |
| 2. | _____ | _____ |
| 3. | _____ | _____ |
| 4. | _____ | _____ |
| 5. | _____ | _____ |
| 6. | _____ | _____ |
| 7. | _____ | _____ |
| 8. | _____ | _____ |
| 9. | _____ | _____ |
| 10. | _____ | _____ |

QUESTIONS

1. What is the role of water in living systems?

2. Why is there a variation of water content in tissues in the materials tested?

3. What is meant by the terms "bound water" and "free water"?

4. Why do solutes come out of water solutions when they freeze?

## EXERCISE 6 - COACERVATES AND EMULSIONS

### GENERAL OBJECTIVE:

1. To develop a concept of how chemicals in the primordial seas could have interacted to form a protoplasm-like substance.

2. To develop the capacity to make valid inferences from observations of experimental data.

### BEHAVIORAL OBJECTIVES:

Upon completion of this exercise, students should be able to:

1. Sketch two or three of the coacervates that were seen in the microscope.

2. Discuss the materials used in this experiment to make coacervates comparable to ones that might have been present in the ancient oceans.

3. Explain what is meant by the isoelectric point of a protein and the properties of proteins at their iso-electric points.

### INTRODUCTION

Scientific investigator A.I. Oparin has established that coacervates are "...bits of semi-liquid colloidal gels, that become separated..." The importance of this is the possibility that in ancient seas, this may have occurred with microscopic particles or organic molecules, which were subsequently surrounded by a membrane. Further postulation establishes one explanation for the evolution of a cellular system. The study of this mechanism is of major importance in the biological laboratory.

## MATERIALS AND EQUIPMENT

microscope and lamp
microscope slides and coverglasses
1 10 ml. pipette or 5 ml. pipette
1 medicine dropper
5% gum arabic (a polysaccharide) in water
test tube with stopper and rack
corn or other vegetable oil
0.1 N HCl
5% gelatin (a protein) in water
gum acacia
mortar and pestle
1 100 ml graduated cylinder
fat stain such as Sudan III
2% $Ca(OH)_2$
acetone
skimmed milk
butter
soapy solution
pHydrion paper

## PROCEDURE

### Part A. Coacervates

1. Mix together in the test tube 5 ml. of 5% gelatin and 3 ml. of 5% gum arabic.

2. Determine the pH of this mixture using pHydrion paper. Place a drop on a microscope slide, cover, and observe under the microscope.

3. Carefully add 0.1N HCl, drop by drop to the test tube. After each addition of acid, mix by inverting the tube and then wait a few seconds to see if the mixture becomes cloudy. If the liquid in the tube remains clear, add another drop of acid. Continue adding acid a drop at a time and inverting the tube until the mixture becomes cloudy.

4. When the material in the tube turns cloudy, make another pH determination. Then, handling the mixture gently, observe a drop under the microscope for the presence of coacervates (which are small spherical droplets). If you do not observe any, decrease the light or go to a higher microscopic power. If you still do not see any coacervates then repeat the whole procedure from the beginning. You may have added the acid too rapidly. When you are successful, record your observations and make sketches of the coacervate droplets.

5. Now add more acid to the test tube, a drop at a time. When the liquid becomes clear again, examine a drop under the microscope and determine the new pH.

Part B. Emulsions

1. Wash a clean mortar and pestle with acetone to remove all traces of grease. Then dry.

2. Grind 12.5 grams of gum acacia to a powder.

3. Add 50 ml. of corn oil (or other vegetable oil). Mix about 2-4 seconds to disperse the oil.

4. Immediately add 25 ml. of water to the mixture. Whip rapidly until thick enough to crackle, then dilute with water to 100 ml. (approximately).

5. Remove a small quantity of the emulsion to a watch glass and add a drop or two of Sudan III - enough to give good color but not too intense. Smear a thin layer on a microscope slide and examine it. The dye colors the oil. Which is the dispersed phase, the oil or the water?

6. Add a few drops of fat stain to a small quantity of skimmed milk. Make a smear and examine under the microscope. Which is the dispersed phase, the oil or the water?

7. Color a small amount of butter with the fat stain. Examine under the microscope. Which is the dispersed phase, the oil or the water?

8. Color a small amount of soapy solution with fat stain and examine under the microscope.

9. Place some of the emulsion (Step 5) in a beaker and heat over a small flame. Pour into a test tube and let stand until cool. Then examine under the microscope as in Step 5.

REFERENCE

Oparin, A.I., A.G. Psynskii, A.E. Braunshtein, T.E. Pavlovokaza, 1959. The Origin of Life on the Earth, MacMillan, New York.

## REPORT SHEET FOR EXERCISE 6

### Part A. Coacervates

1. Sketch two or three of the coacervates that you see in the microscope.

2. Discuss in your group how the materials you used in this experiment to make coacervates compare with the ones that might have been present in the ancient oceans.

3. What is meant by the isoelectric point of a protein and what properties of proteins are exhibited at their isoelectric points?

### Part B. Emulsions

Complete the following chart.

| Preparation | Dispersed Phase | Continuous Phase |
|---|---|---|
| oil emulsion | oil | water |
| skimmed milk | | |
| butter | | |
| soapy solution | | |

QUESTIONS

1. What happened to heated and cooled oil emulsion?

2. What is the function of the gum arabic?

3. What is a colloid?

## EXERCISE 7 - THE PHYLA OF THE ANIMAL AND PLANT KINGDOMS

### GENERAL OBJECTIVE:

1. To become familiar with the structural characteristics common to certain phyla.

2. To develop a concept of classification.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students should be able to:

1. List five structural differences between organisms of the plant and animal kingdoms.

2. Point out some of the structural differences between organisms of the same phylum.

3. Indicate the differences between several of the common phyla listed in Charts I & II.

4. Cite, orally, some beneficial aspects of several of the commonly overlooked organisms.

### INTRODUCTION

Biologists utilize structural similarities and diverse characteristics of all living things as the basis for coordinating a unifying scheme of classification. These structural similarities and diversities begin with major, broad divisions and conclude with specific groups or species, or begin with subdivisions and conclude with specific groups or species, or may even conclude with subdivisions of the species (such as varieties and strains). Each hierarchical division in descending order is a subdivision of the preceding division and shows greater specificity. This identification process is comparable to reading the name of the addressee on an envelope in reverse order. One ends with the name of the individual member of a given family to whom the letter is addressed.

In this exercise you will examine representatives of the various phyla of living organisms and observe those characteristics by which they are classified.

## MATERIALS AND EQUIPMENT

Bring living specimens of both domestic and captured harmless animals, and plants from their respective soil and water habitats. Several species of all phyla should be represented.

| | |
|---|---|
| Microscope | Stereoscopic microscope |
| Microscope slides and coverslips | Dropping pipette |

## PROCEDURE

Examine each specimen with the necessary optical instruments provided, if these are required; otherwise examine with the unaided eye.

The students should bring one or more references to class to be used in conjunction with their examination of the various organisms. Examine each specimen displayed and fill in the chart on the report sheet.

A demonstration microscope may be prepared by the instructor to show stained slides of bacteria or living motile bacteria in a hanging drop mount.

A. Animal Classification - Chart I

B. Plant Classification - Chart II

## REFERENCES


Alexander, Taylor R., R. Will Burnett, Herbert S. Zim, 1970. Botany, Golden Press, New York.

Berril, N.J., 1966. Biology in Action, Dodd, Mead and Company, New York.

Bold, Harold C., 1964. The Plant Kingdom, Prentice-Hall, Inc. Englewood Cliffs, New Jersey.

Burnett, R. Will, Harvey I. Fisher, Herbert S. Zim, 1958. Zoology - An Introduction to the Animal Kingdom, Golden Press, New York.

Laubengayer, Richard, Willis H. Johnson, Louis E. Delanney, and Thomas A. Cole, 1966. Biology, Holt, Rinehart and Winston, New York.

McElroy, William D., Carol O. Swanson, Neal D. Buffaloe, Arthur W. Golston, and Robert L. Macey, 1968. Foundations of Biology, Prentice-Hall, Inc., Englewood Cliffs, New Jersey.

Schers, Robert W., 1963. Plants for Man, Prentice-Hall, Inc., Englewood Cliffs, New Jersey.

Simpson, George Gaylord and William S. Beck, 1969. Life - An Introduction to Biology, Harcourt, Brace and World, Inc., New York.

## REPORT SHEET FOR EXERCISE 7 - Charts 1 and 2

### CHART I

*The Animal Kingdom

| Phylum | Name of Organism Observed | **<br>1. unicellular or<br>2. multicellular | Outer Covering<br>1. hair or fur<br>2. feathers<br>3. scales<br>4. none | Skeleton<br>1. absent<br>2. internal<br>3. external | Limbs<br>1. absent<br>2. present | Respiratory Organs<br>1. gills<br>2. lungs<br>3. others | Special characteristics of phylum or class |
|---|---|---|---|---|---|---|---|
| Protozoa | | | | | | | |
| Porifera | | | | | | | |
| Coelenterata | | | | | | | |
| Playthelminthes | | | | | | | |
| Nematoda | | | | | | | |
| Mollusca | | | | | | | |
| Annelida | | | | | | | |
| Arthropoda | | | | | | | |
| Echinodermata | | | | | | | |
| Chordata | | | | | | | |
| Subphylum Vertebrata | | | | | | | |
| Class Agnatha | | | | | | | |
| Class Chondrichthyes | | | | | | | |
| Class Osteichthyes | | | | | | | |
| Class Amphibia | | | | | | | |
| Class Reptilia | | | | | | | |
| Class Aves | | | | | | | |
| Class Mammalia | | | | | | | |

*Although several other systems of classification exist which subdivide the conventional taxonomical kingdoms into several additional kingdoms, namely, Protista and Monera, this scheme has been chosen because of convenience. The teacher may alter it to accommodate the availability of materials and references.

**Indicate by writing the correct numbers under the headings

## CHART II

*The Plant Kingdom

| Phylum | Name of Organism Observed | 1. unicellular or 2. multicellular | Pigmentation 1. absent 2. present | Seeds 1. absent 2. present | Characteristics of Division Subdivision, or Class |
|---|---|---|---|---|---|
| A. Division Thallophyta 1. Subdivision Algae including blue-green algae | | | | | |
| 2. Subdivision fungi including bacteria | | | | | |
| B. Division Bryophyta | | | | | |
| C. Division Pteridophyta | | | | | |
| D. Division Spermatophyta | | | | | |
| 1. Subdivision Gymnospermae | | | | | |
| 2. Subdivision Angiospermae | | | | | |
| a. Class Dicotyledonae | | | | | |
| b. Class Monocotyledonae | | | | | |

*Although several other systems of classification exist which subdivide the conventional taxonomical kingdoms into several additional kingdoms, namely, Protista and Monera, this scheme has been chosen because of convenience. The teacher may alter it to accommodate his availability of materials and references.

## QUESTIONS

1. Locate 10 different plants and animals in your neighborhood, campus or community environment and classify them according to Kingdom, Phylum, and, if chordates, Class of subphylum vertebrata.

2. Compare the appendages (limbs) of representatives of the classes of vertebrates examined and note how they are adapted to the habitat and mode of locomotion of those organisms.

3. Are all seed plants flowering plants? Explain and cite examples.

4. Of what value are the algaē:

   a. Economically?

   b. In global ecology?

5. What are the seven common taxonomic ranks used to classify organisms?

6. What is the difference between the taxonomic categories "phylum" and "division"?.

7. What are the Kingdom, Phylum, Class, Order, Family, Genus and Species into which human beings are grouped?

8. What characteristics enable one to place human beings in their appropriate Kingdom, Phylum, Class, Order, Family, Genus or Species?

## EXERCISE 8 - DNA REPLICATION AND TRANSCRIPTION BY mRNA AND tRNA TO SEQUENCE A PROTEIN

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GENERAL OBJECTIVE:

1. To develop an understanding of how proteins are formed.

2. To develop an understanding of how symbols can be used to explain the way in which genes convey messages about the formation of proteins.

BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students should be able to:

1. List the five types of bases in DNA and RNA.

2. Identify a letter coded in units of three bases from DNA and RNA.

3. Identify the complementary base pairs in a strand of DNA and the corresponding bases in mRNA and tRNA.

4. Decode a series of given DNA sequences and show how the genetic code can be used as a language for communicating ordinary linguistic information.

INTRODUCTION

DNA is a coded chain of nucleic acids. The duplication and translation of this code is necessary for the body to make proteins. There are about three or four steps involved in protein synthesis.

This exercise is designed to help you understand how DNA is replicated, transcribed and then translated into amino acids.

Proteins are made up of chains of amino acids. Each amino acid is encoded by at least a triplet of nucleic acid bases in the DNA structure. The copying (transcription) of the codes in the DNA takes the form of messenger RNA (mRNA). The decoding is done by the transfer RNA (tRNA). Each amino acid is coded by the tRNA.

PROCEDURE

The code letters for one DNA strand are given below. Spaces are provided for transcribing the DNA strand into the mRNA and tRNA. The amino acids associated with these tRNA sequences are given in the table on the next page. The game alphabet equivalent is supplied in the same table.

Example 1. Work out the message on the DNA strand sequence given below.

| Example: | CAC | ACG | CAC | CTG | ATC | GTA | CAA | ATA | ACA | GTA |
|---|---|---|---|---|---|---|---|---|---|---|
| Answer: | | | | | | | | | | |
| DNA Strand: | CAC | ACG | CAC | CTG | ATC | GTA | CAA | ATA | ACA | GTA |
| mRNA Codons: | GUG | UGC | GUG | GAC | UAG | CAU | GUU | UAU | UGU | CAU |
| tRNA Nodocs: | CAC | ACG | CAC | CUG | AUC | GUA | CAA | AUA | ACA | GUA |
| Amino acids: | His | Thr | His | Leu | Heu | Val | Glu | Heu | Thr | Val |
| Message: | P | E | P | S | I | N | O | G | E | N |

Example 2. Work out the base sequence on strand 2 of the given DNA strand above as:

| Given Strand 1: | CAC | ACG | CAC | GTG | ATC | GTA | CAA | ATA | ACA | GTA |
|---|---|---|---|---|---|---|---|---|---|---|
| DNA Strand 2: | GTG | TGC | GTG | gac | tag | cat | gtt | tat | tgt | cat |

Key: A = Adenine C - Cytosine
G = Guanine T = Thymine
U = Uracil

Nodocs and Alphabetical Game Equivalents of Amino Acids

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| AAA | Lys | A | CGA | Arg | Q | UAA | Space | |
| AAG | Lys | A | CGG | Arg | Q | UAG | Comma | |
| AAC | Aspn | B | CGC | Arg | Q | UAC | Tyr | Period |
| AAU | Aspn | B | CGU | Arg | Q | UAU | Tyr | Question |
| AGA | Arg | C | CCA | Pro | R | UGA | Try | U |
| AGG | Arg | C | CCG | Pro | R | UGG | Try | U |
| AGC | Ser | D | CCC | Pro | R | UGC | Cys | V |
| AGU | Ser | D | CCU | Pro | R | UGU | Cys | V |
| ACA | Thr | E | CUA | Leu | S | UCA | Ser | W |
| ACG | Thr | E | CUG | Leu | S | UCG | Ser | W |
| ACC | Thr | F | CUC | Leu | T | UCC | Ser | X |
| ACU | Thr | F | CUU | Leu | T | UCU | Ser | X |
| AUA | ILEU | G | GAA | Glu | J | UUA | Leu | Y |
| AUG | Met | H | GAG | Glu | J | UUG | Leu | Y |
| AUC | ILEU | I | GAC | Asp | K | UUC | Phe | Z |
| AUU | ILEU | I | GAU | Asp | K | UUU | Phe | Z |
| CAA | Glu | O | GCA | Ala | L | | | |
| CAG | Glu | O | GCG | Ala | L | | | |
| CAC | His | P | GCC | Ala | M | | | |
| CAU | His | P | GCU | Ala | M | | | |
| | | | GUA | Val | N | | | |
| | | | GUG | Val | N | | | |
| | | | GUC | Val | N | | | |
| | | | GUU | Val | N | | | |

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## REPORT SHEET FOR EXCERISE 8

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**Strand One DNA Sequences To Be Decoded**

DNA Strand 1: GCA ACA CTT TAA CTC ATG ACG CCC ACG TAA
AAT ACA TAA GCA
DNA Strand 2:
mRNA Codons:
+RNA Nodocs:
Amino Acids:
Amino Acids:
Message:

DNA Strand 1: GAC GTT CAA TCG GCG AGC ATA ACA TAA
ATT CTG TAA
DNA Strand 2:
mRNA
tRNA
Amino Acids:

Message:
Amino Acids:
+RNA Nodocs:
mRNA Codons:
DNA Strand 1: ATC ACT ACA TAC
DNA Strand 2:

Message:
Amino Acids:
+RNA Nodocs:
mRNA Codons:
DNA Strand 1: TCA ATG AAA CTT TAA ATG AAG CTT ATG TAA
ATA CAG AGT TAA TCG
DNA Strand 2:

Message:
Amino Acids:
rRNA Nodocs:
mRNA Codons:
DNA Strand 1: CCA CAA TGG ATA ATG CTT TAT
DNA Strand 2:

Message:
Amino Acids:
+RNA Nodocs:
mRNA Codons:
DNA Strand 1: CTA CTT TGA AGT TTA TAA GTA AAA CTC
TGA CCG ACA TAA GTC

DNA Strand 2:

Message:
Amino Acids:
+RNA Nodocs:
mRNA Codons:
DNA Strand 1: CAG CTT TAA AAC CAA CAG
GAT CTA TAA CAA GTC GCG
TTA
DNA Strand 2:

Message:
Amino Acids:
tRNA Nodocs:
mRNA Codons:
DNA Strand 1: CAT CAG TCA ACA CCA TAT
DNA Strand 2:

Message
Amino Acids:
tRNA Nodocs:
mRNA Codons:
DNA Strand 1: GCC AAG GAC ACA TAA ATG AAA
CTG CTT ACG
DNA Strand 2:

Message:
Amino Acids
tRNA Nodocs:
mRNA Codons:
DNA Strand 1: GCT AAA GAT ACA TAA CTA CAA GCC ACG TAA
DNA Strand 2:

Message:
Amino Acids:
tRNA Nodocs:
mRNA Codons:
DNA Strand 1: CTG AAG CCT CTG AAG CTA CAC
AAA CCA ATC GCA GCG AAA TAC
DNA Strand 2:

Message:
Amino Acids:
tRNA Nodocs:
mRNA Codons:
DNA Strand 1: GCG ACA CTC TAA TGA CTG TAA GCT AAA
GAT ACA TAA TGA CTA
DNA Strand 2:

Message:
Amino Acids:
tRNA Nodocs:
mRNA Codons:
DNA Strand 1: TAA AAA TAA GCC AAG GTT TAC
DNA Strand 2:

QUESTIONS

1. What are some of the differences between DNA and RNA?

2. What effects will mutation have on the protein structure that is coded?

3. What is a gene?

4. Why don't all cells of an organism produce all the proteins that are coded in the genetic materials?

5. Why do certain specific cells produce only certain specific proteins, i.e., hemoglobin by red blood cells, keratin by epidermal cells?

## EXERCISE 9 - THE INHERITANCE OF GENETIC TRAITS

### GENERAL OBJECTIVES:

1. To develop an understanding of Mendelian genetics.
2. To develop skill in the use of the Punnett Square technique for predicting types of offspring when the genetic make-up of parents is known.
3. To develop problem-solving skills in genetics.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, the student should be able to:

1. Explain the difference between phenotype and genotype.
2. Employ the Punnett Square technique to predict the genotype(s) in the offspring from the monohybrid cross Tt x Tt.
3. Compare and contrast the following terms:
   a. dominant and recessive traits.
   b. heterozygous and homozygous alleles.
   c. homozygous dominant and homozygous recessive.
   d. complete and incomplete dominance.

### INTRODUCTION

#### Part A. The Inheritance of Starch and Color in Corn Seeds

Studying inheritance in corn grains using Mendelian principles has certain basic advantages. One of these advantages is the ease with which observations can be made because the grains are held in place on the ear of corn, which makes the counting of individuals easy. Each grain of corn you will use represents a separate offspring of a cross between the female gametophyte and the male gametophyte. The sperm cells carried within pollen grains fertilize the egg cells situated within the ovules, which then develop into seeds.

In producing ears for the first cross you will study, the plant-breeder selected parents that were pure-breeding (homozygous). Two pure strains-tall and short TT x tt) were crossed; they produced the $F_1$ offspring plants whose genotype is Tt. These Tt plants were next crossed with each other (selfed), producing the $F_2$ generation.

The second kind of cross you will study involved an $F_1$ parent (Tt) and the recessive parent plant (tt), producing a backcross generation.

## PROCEDURE

Work in teams of three students (one counter and two tabulators). Each team will receive an ear of corn. Examine the ear of corn, classify the kernels, and record phenotypic characters as follows:

a. starchy (smooth)

b. sweet (shrunken)

c. waxy

d. purple

e. yellow

Caution: Be careful when handling the ears, or the kernels will become loose and drop out. Do not pick the kernels from the ears!

Record the characters on the report sheet in the column titled "Tally" by making one mark for each type of kernel. To determine the number of kernels, place a pin in the ear at the beginning of the row of kernels where counting starts. Determine the phenotypic character of each kernel in each row until you return to the starting point marked by the pin. As the characters are determined, tally each character in the appropriate row in the "Tally" column report sheet.

Total the number of each type of kernel on your ear of corn and place this number in the "Totals" column. Calculate the percentage of each type. Are the percentages close to any ratio of small whole numbers? If so, what? Is your ear of corn an $F_2$ generation or a backcross generation? From your

tabulations, can you determine the probable strain of the $F_1$ corn? The probable strain of the parent ears? What trait is dominant? What trait is recessive? Show by using Punnett Squares the probable cross which produced the progeny represented by your ear of corn. Write a clear statement for each of the Mendelian principles illustrated.

## Part B. Some Problems in Genetics

Dominant and Recessive Traits

1. In peas, the gene for tallness (T) is dominant to the gene for shortness (t). What offspring phenotypes would be expected from the following crosses, and in what proportions?

   a. heterozygous (Tt) x heterozygous

   b. heterozygous x homozygous tall (TT)

   c. homozygous tall x homozygous short (tt)

2. If blue eye color in man is recessive to other colors, could

   a. brown-eyed parents have a blue-eyed child?

   b. blue-eyed parents have a brown-eyed child?

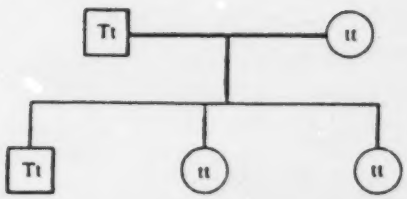
3. Albinism in plants (the inability to synthesize chlorophyll) is a recessive character in a number of plant species. If a tobacco plant known to be heterozygous for albinism is self-pollinated and 600 of its seeds are subsequently germinated:

   a. how many seedlings would be expected to be albino?

   b. how many would be expected to have the parental genotype?

4. A man and his wife can both taste phenylthiourea. They have four children, two of whom are unable to taste it. What are the parental genotypes?

5. Construct pedigree diagrams for the following families, giving the possible genotypes for each member. Represent males with a square and females with a circle, and write the genotype along-side or inside the symbol. Example: A man who is a taster has a non-taster wife; they have a taster son and two non-taster daughters.




a. A man and his wife are both tasters and they have two taster sons, a taster daughter, and a non-taster daughter.

b. A taster man has a taster son, a non-taster son, and a taster daughter by his first marriage and two non-taster daughters by his second marriage. His first wife was never tested but his second wife was a non-taster.

c. The daughter of the first marriage in b above subsequently married a non-taster and produced eight children, all of whom were tasters.

6. When Poggy, a colored male hamster, was mated with Zoe, a white female, the offspring were all colored. When Zoe was later mated with Prince, who was colored, some of this second litter were white.

a. What might one conclude about the genotypes of Poggy, Prince, and Zoe with respect to coat color?

b. If an animal of the first litter were to be mated with a colored animal of the second litter, what offspring phenotypic ratios might be expected?

c. What offspring might be expected from a cross between an animal of the first litter and a white animal from the second litter?

The 1:2:1 Phenotypic Ratio

7. A farmer crossed his roan bull with three groups of cows. Cows of Group A were white; cows of Group B were red; and cows of Group C were roan. What proportions of the different coat colors would be expected in the offspring of each group of cows?

8. If the same farmer decided to make his Shorthorn herd exclusively roan coat by selling all white and red calves, would the color of the bull that he used make any difference economically?

Observed and Expected Ratios

9. In an investigation of the nature of length of leg in domestic fowl the following two crosses were made and the number of offspring recorded:

| | Number of Short Offspring | Number of Normal Offspring |
|---|---|---|
| 1. normal x short leg | 48 | 51 |
| 2. short leg x short leg | 55 | 28 |

From these figures, what might be concluded about the action of this gene?

10. Brachyphalangy in humans is a condition in which the middle bone of the fingers is greatly shortened and usually fused to the next bone, with the result that the fingers appear to have only two joints. A brachyphalangic person is heterozygous for a lethal gene; the homozygous dominant offspring is born without digits and with gross skeletal defects which soon cause death.

    a. In a marriage between a brachyphalangic man and a normal woman what proportion of their children would be expected to be brachyphalangic?

    b. Show this diagrammatically.

    c. What ratio of brachyphalangic to normal children would be expected in a family in which both parents are brachyphalangic?

    d. Show this diagrammatically.

Phenotypic Ratio Involving Two Pairs of Genes: The 9:3:3:1 Ratio

11. If a maize plant heterozygous for the alleles for pigmy and crinkly-leaf (both recessive to normal size of plant and normal leaf) is self-pollinated and 160 of the seeds are subsequently collected and germinated, how many would you expect to show:

    a. crinkly leaves?

    b. normal size?

    c. normal leaves and normal size?

    d. normal leaves and pigmy?

12. In a certain breed of domestic fowl, pea comb is dominant to single comb, but feather color shows absence of dominance: black feathers and white feathers are homozygous, e.g., BB and bb respectively, whereas the heterozygote Bb gives "blue" feathers.

From crosses between birds heterozygous for both alleles, what proportion of the offspring would be expected to be:

    a. pea combed?

    b. black feathered?

    c. blue feathered?

    d. pea combed and blue feathered?

    e. single combed and white?

13. In Drosophila, curved wing (c) and forked bristle (f) are recessive to the normal or wild type wing and bristle shape (expressed as + in each case). Show diagrammatically, the following crosses:

    a. +c +f x cc ff;

    b. ++ +f x +c ff;

    c. +c +f x +c ff.

14. What proportion of the offspring from the cross in question 13c would be expected to show the wild-type phenotypes for both alleles?

15. In fowl, two pairs of alleles (Rr and Pp) regulate the shape of comb. Two cocks, each of which has a pea-shaped comb, are mated to five hens as follows:

| Cock X x | Hen | Comb | Offspring | comb ratios |
|---|---|---|---|---|
| | A | walnut | 3 walnut:<br>1 single | 3 pea: 1 rose: |
| | B | pea | 3 pea: | 1 single |
| | C | rose | 1 walnut:<br>1 single | 1 pea: 1 rose |
| | D | single | all pea | |
| | E | rose | all walnut | |

What simple genetic explanation accounts for the inheritance of comb shape, and what are the genotypes of the cocks and hens above?

16. In peas the gene for round seed(R)is dominant to wrinkled (r), and yellow seed coat (Y) is dominant to green (y). Consider a cross between peas heterozygous for both alleles:

    a. What proportion of the seeds produced would be expected to be wrinkled?

    b. What proportion would be expected to be yellow?

    c. What proportion would be expected to contain a homozygous pair of alleles?

17. In humans, a recessive gene for a severe disease of the blood vessels is heterozygous in about 0.05% of the population. A man whose brother died from the disease is contemplating marriage and consults a genetics counselor because he is worried about the chances of a child of his having the disease. What advice might the counselor give?

18. If the man in question 17 marries his first cousin what are his chances of having a child with the disease?

19. What is the probability that a family of four children will consist of

    a. four boys?

    b. three boys and a girl?

    c. two boys and two girls?

    d. one boy and three girls?

    e. four girls?

    What is the connection between this question and the previous one?

20. When pink sweet peas were self-pollinated and the seeds were collected and sown, the following flower colors were obtained:

    red 36 pink 56 white 28

    Are these results consistent with the hypothesis that pink flowers are heterozygous for a single pair of color alleles, showing absence of dominance?

Sex Chromosomes and Sex-Linkage

21. If a sex-linked gene is a recessive lethal, causing death and reabsorption of the embryo at an early stage,

    a. What proportion of the offspring would be expected to be females?

    b. What proportion of the offspring would be expected to be carriers of the gene from a mating between two mammals, the female of which carries the gene in its heterozygous state?

22. In cats, a coat color gene is located on the non-homologous portion of the X chromosome. BB or B- is black, bb is yellow, and Bb is tortoiseshell. Decide if the following statements are true or false, and justify your answers.

    a. A mating of a black tom with a yellow tabby could give female tortoiseshell kittens.

    b. The same mating can give male tortoiseshell kittens.

c. A yellow male mated with a tortoiseshell female could give black male kittens.

d. The same mating could give black female kittens.

e. A tortoiseshell tabby can have tortoiseshell kittens by whatever color tomcat with whom she mates.

23. A normal-sighted woman whose father was red-green colorblind married a man with normal vision. What ratio of genotypes and phenotypes would be expected among their children?

24. The gene for the ability to taste PTU is autosomal and dominant to non-tasting. Red/green colorblindness is sex-linked and recessive.

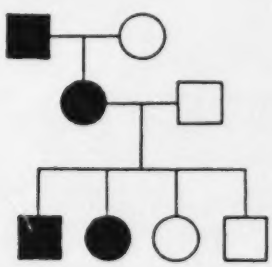
A man and his wife, both of whom can taste PTU and have normal vision, have a child who is colorblind and a non-taster.

a. What is the child's sex?

b. What is the probability (expected occurrence) of this child's phenotype in their subsequent children?

c. What is the probability that the child will be a taster with normal vision, like its parents?

25. Consider the following pedigree covering three generations of a mammal. The symbol • depicts a female who shows a certain trait in the phenotype, and □ represents a normal male.



a. Does this show evidence of sex-linkage?

b. Under what conditions could this pedigree represent the inheritance of a sex-linked gene?

c. Can a father and his son both be hemophiliacs?

Skin Color In Humans: Multiple Genes

26. On the basis of Davenport's findings that skin color in humans depends on the pigment genes present on two unlike loci (two pairs of genes), and that white skin can be represented as aaaa or $a^4$, how would you describe the following:

a. black skin genotype?

b. mulatto?

c. dark mulatto?

d. light mulatto?

27. Show algebraically, the cross between

a. Two dark mulattos:

b. Two light mulattos;

c. One light mulatto and one dark mulatto.

For each, show the algebraic expression that represents the possible genotypes of their offspring.

28. a. What proportion of the children of a marriage between two light mulatto parents would be expected to have white skins?

b. What proportions would be expected to have skin the same color as their parents?

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## REPORT SHEET FOR EXERCISE 9

Part A. Inheritance of Starch and Color in Corn Seeds

| CHARACTERS | TALLY | TOTALS |
|---|---|---|
| Starchy (smooth) | | |
| Sweet (wrinkled) | | |
| Waxy | | |
| Yellow | | |
| Purple | | |

Ratio of starchy to sweet is:

Ratio of purple to yellow is:

Is your ear of corn in the $F_1$ or $F_2$ generation?

Which of the above traits are dominant and which recessive?

Form a Punnett Square for the possible genotypes producing the phenotypes observed.

What Mendelian principles are illustrated?

Questions

1. Write a statement which explains the difference between ratio and percentage.

2. Write a statement which explains the relationship between genes and chromosomes, and between genes and loci.

3. Discuss the relationship of genes to chromosomes.

4. Discuss the concept of sex-linkage.

5. Write a statement indicating the distinction between sex chromosomes and autosomes.

6. Distinguish a Punnett Square from a pedigree chart.

7. Illustrate how Mendelian principles apply in relation to varieties, or species, in the natural environment.

## EXERCISE 10 - THE INHERITANCE OF SPECIFIC PROTEINS

### GENERAL OBJECTIVES:

1. To be aware of blood typing procedures.
2. To perceive the morphological and physiological nature of blood.
3. To consider the significance of sickle cell disease as a genetic disease.
4. To become proficient in predicting certain phenotypes and genotypes.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students should be able to:

1. Distinguish between "antigen" and "antibody".
2. Differentiate between gene frequencies and gene ratios.
3. Determine blood types in a laboratory.
4. Perform a test for sickling of red blood cells.
5. Predict the phenotypes and genotypes of offspring when given certain parental genotypes.

### INTRODUCTION

Bloodletting was a medical practice that extended from antiquity into the 20th Century. The medicinal leech (Hirudo medicinalis) could be purchased in drug stores in the United States as late as the 1920's. At the turn of the century, some physicians began to wonder if bleeding the sick might be a cause of death for many of their patients. They reasoned that it might be helpful to give a patient blood instead of taking it away from him.

The first of these tissue transplants (blood being a connective tissue) was successful but some of those that followed were not. Through the pioneering work of Moss, Jensky and Landsteiner (working independently), it was discovered that something in the blood plasma was causing some of the donated blood to clump (or agglutinate). They, not knowing what factor was causing this effect, called it an "antibody". The term antibody was coined 50 years earlier by Louis Pasteur.

We have since learned that blood contains proteins. Among these proteins are those called "antigens". Antigens are a part of almost all proteins and some of the more complex carbohydrates and nucleic acids. There are different types of antigens. Thus one person may possess antigen "A" while another person may have "B", or a combination of A and B (AB). Some individuals are known not to possess these specific antigens (A, B, or AB) in their blood, and are therefore said to have "O" type blood. Antigens are capable of stimulating the body to produce another form of proteins (gamma globulins) called "antibodies". Antibodies are produced by plasma cells which are parts of the blood system of higher vertebrates.

Antibodies are specific and therefore each type is usually produced in response to a specific protein. If the structure of the protein (antigen) changes, the original antibody will become less able to react to it and therefore a new type of antibody will have to be produced.

Since we can stimulate the production of antibody proteins in a number of animals (rabbits, horses, etc.) by injecting them with the antigen protein we wish to identify, it becomes a very convenient tool for locating specific proteins without the longer process of physical-chemical analysis.

Humans form a few antibodies spontaneously. One example of this is the antibody formed against Pneumococcus type 7. Another form is the production of antibodies against the kind of cell membrane protein (A or B) which is not made by the recipient. The antibody protein against protein A is called "anti-A" or "alpha". The one against protein B is called "anti-B" or "beta". People who form protein A spontaneously form an anti-B or beta antibody. Those that form protein B spontaneously form an anti-A or alpha antibody. The formation of antibodies was a puzzlement to Drs. Moss, Jansky and Landsteiner.

About 85% of the white population in the United States, and a slightly higher percentage among Blacks and Orientals, form another group of membrane proteins. These are designated C, D and E, and constitute the Rhesus Monkey Factor (Rh factor). Proteins A and B are also found among other Primates. Antibodies against the Rh factor are not formed spontaneously but are formed if Rh protein is infused into the blood system of an Rh-negative person, that is, one who does not form Rh protein.

A considerable list of proteins in cell membranes, including red blood corpuscle membranes, has now been identified. The names Kelly, Duffy, Lewis, etc. denote the names of the patients in whom these proteins were first discovered to be missing.

Proteins A, B, C, D, and E are quite different. However, there are some substitutions of a single amino acid into the hemoglobin chains which may have far-reaching effects and are responsible for such blood disorders as thalassemia and sickle-cell anemia. The resulting change is not easily detected with antibodies. Sickle-cell anemia (a homozygous recessive condition) and sickle-cell trait (the heterozygous condition) can be detected by a simple test.

The genes $I^A$ and $I^B$ which regulate the production of proteins A and B are not dominant to each other. So, a person who has both genes $I^A$ and $I^B$ will produce both proteins A and B and therefore be of type AB. A person who is homozygously recessive for the genes ($I^a$ and $I^b$) will form neither protein A nor B and is designated as type O. Genes $I^A$ and $I^B$ are dominant to their recessives, so that a person of type A can be either homozygous ($I^A$ $I^A$) or Heterozygous ($I^A$ $I^B$).

| | | | |
|---|---|---|---|
| Type A | $I^AI^AI^bI^b$ | or | $I^AI^aI^bI^b$ |
| Type B | $I^aI^aI^BI^B$ | or | $I^AI^aI^BI^b$ |
| Type AB | $I^AI^AI^BI^B$ | or | $I^AI^aI^BI^B$ or $I^AI^AI^BI^bI$ or $I^AI^aI^BI^b$ |
| Type O | $I^aI^aI^bI^b$ | | |

There are about 3 billion combinations for the known blood cell proteins, indicating that each person on earth probably has a protein "fingerprint".

## MATERIALS AND EQUIPMENT

Anti-A blood serum
Anti-B blood serum
Sodium sulfite (20%)
Ethyl alcohol (70%)
Normal saline solution (.8990)
Cotton swabs (disposable)
Sterile blood lancets (disposable)
Grease pencils
Microscope slides
Paper towels
Light microscopes

PROCEDURES

Preparations

1. Dry a clean slide from the alcohol bath using a lint-free cloth or paper towel. Draw two circles about 3/4 inch in diameter with a wax pencil. Label the left circle A and the right one B. Put your initials on the end of the slide.

2. Place about 1 ml (1 inch) of normal (0.89%) NaCl in a small (7mm.) test tube and place the tube in a rack or other support.

3. Wash your hands if you are the subject. Wipe the finger pad of the third digit on the left hand (if you are righthanded) with some 70% alcohol on a pledget of cotton. Next wipe the fingertip with ether on a pledget of cotton. The evaporation of the ether helps to rapidly remove the alcohol (which will destroy red blood corpuscles), at the same time providing a cooling effect which draws blood into the fingertip.

Obtaining a Few Drops of Fresh Blood

4. Have your partner open a packet containing a sterile disposable, metal lancet, vis:

5. Place the left hand palm upward on the table with the cleaned fingertip extended. Have your partner prick the ball of the fingertip with the lancet. The skin at this point is about 1/4 inch thick so the lancet should go all the way to the stop device. Remove and discard the lancet.

6. Two or three drops of blood can be made to flow out by gentle pressure on the fingertip. Add this blood to the tube, shaking to mix. Make a medium pink suspension. If it is red, add more saline. How will this prevent pseudoagglutination?

CAUTION! Do not touch the test tube with the wounded finger. After blood has been obtained cover the wound with an alcohol-saturated pledget to stop the bleeding and to prevent infection.

Test for Sickling

Mix a drop of blood and a drop of 20% sodium sulfite solution on a microscope slide. Cover with a coverslip and seal with melted petrolatum (petroleum jelly) applied with a small brush. Let stand. Sickle cell disease will cause sickling in about 15 minutes. Sickle cell trait will take about 30 minutes or a little longer.

A Problem in Cross-Matching:

| ABO TYPE | CELL ANTIGEN | PLASMA OR SERUM ANTIBODY |
|---|---|---|
| O | Neither | alpha and beta |
| A | A | beta |
| B | B | alpha |
| AB | A and B | Neither |

Type A or Type B blood can be used to determine any other type if the titer of an antibody is high enough and there are no inhibitors (H in Type O blood). Let us use Type A blood as an example and assume no interference.

Procedure for Obtaining Materials

Separate the cells from plasma by centrifugation or from serum by letting blood samples clot. One sample here is Type A blood. Now mix Type A cells with serum or plasma of the untyped blood and mix Type A serum (containing an Anti-B or beta antibody) with cells of the untyped blood. Mix and let stand. Look for agglutination.

Work out this problem using Type B blood cells and serum to determine unknown types.

| CASE | BLOOD TYPE | TYPE A CELLS AND UNTYPED SERUM | TYPE A SERUM (BETA) AND UNTYPED CELLS |
|---|---|---|---|
| I | B | Clumped B, Ø | Clumped B, A, B |
| II | A | Not Clumped A, A, B | Not Clumped A, Ø |
| III | O | Clumped B, O | Not Clumped A, O |
| IV | AB | Not Clumped A, AB | Clumped B, AB |

NOTE: Where the Untyped Cells are in Type A Serum (beta), one must write down the ABO Type of the blood with (or without) the antibody concerned.

Rh Types (Types C, c, D, d, E, e)

The Rh factors are cell proteins C, D, and E. If one is positive for any of these, the capital letters are used to show its presence as the result of genetic dominance for the formation of these specific proteins. Small letters are used to indicate the absence of these proteins due to genetic recessiveness (Rh Negativity).

1. Take a clean slide from alcohol and dry it with a paper towel. Handle the slide by its edges.

   Draw three 3/4 inch diameter circles, label them C, D, and E. Write your initials on the end of the slide and place it on the Rh-typing box for a few minutes to warm up.

3. Obtain from the teacher a slide with a drop of Anti-C, Anti-D, and Anti-E sera in the proper circles. Add a drop of undiluted blood to each. Mix with separate toothpicks and place on the Rh-typing box. Shake for 4 minutes, observing results at 1 minute intervals.

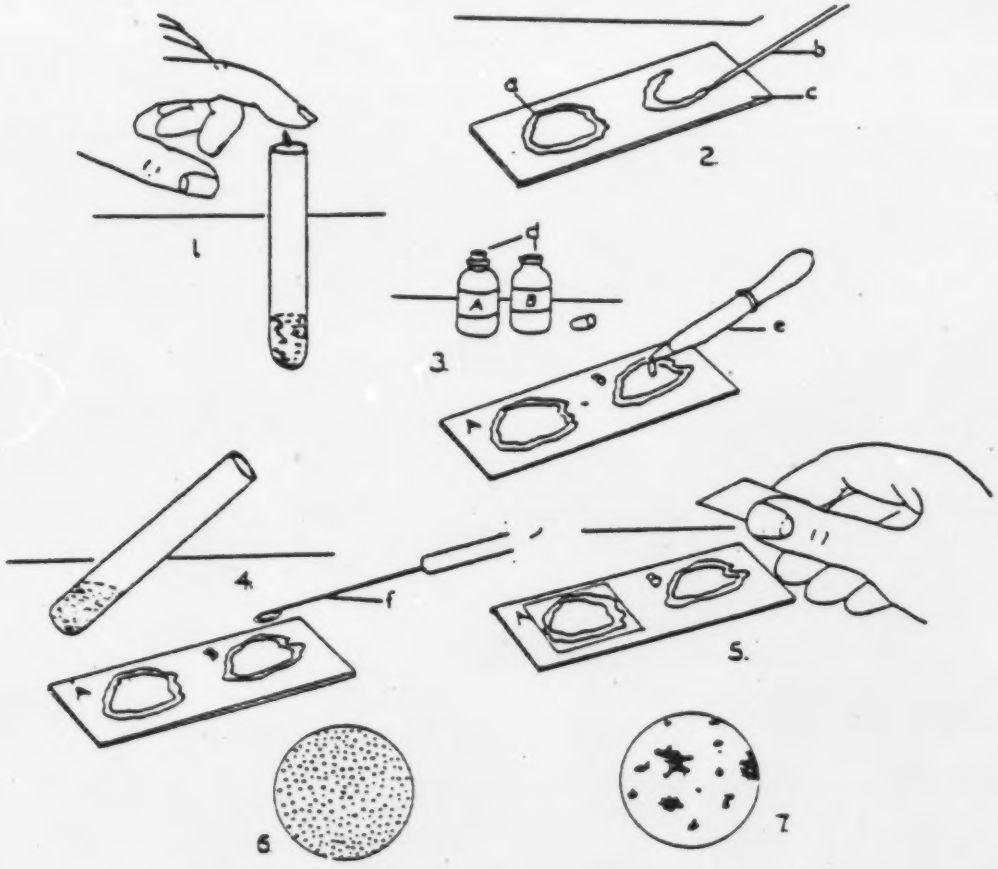
4. Report your results for C, D, and E separately, using capital letters for positive reactions, and small letters for negative ones.

ABO Typing Using Anti-A and Anti-B Serum

Take the slide with marked circles to the teacher's table and obtain a drop of Anti-A serum in circle A and a drop of Anti-B serum in circle B. Return to your workplace with the slide and add a drop of the blood suspension prepared in Step 6 (pp. 21-3). Be careful not to let the dropper touch either of the drops of serum. Mix the contents of each circle by stirring with different ends of a toothpick or an applicator stick.

Examine the preparations under the low power (10X) objective of the microscope. Type A blood will clump on the A side and Type B blood will clump on the B side. Type O will not clump in either circle, but Type AB will clump in both.

## An Alternate Procedure




Open Slide Blood Typing Method. (Illustrating the procedure that may be followed in blood typing).
1—collecting two or three drops of blood in a test tube with a few cc. of normal saline;
2—by means of a vaseline "gun," b, a circle of vaseline is placed on each end of the slide, C;
3—the one circle is labeled A and the other B and a drop of A and B type sera, d, is added to each respectively by means of a small pipette, e;
4—a small drop of diluted blood of unknown type in the test tube is added to each drop with a platinum wire loop (flame well between drops);
5—coverslips are placed over each to prevent evaporation. The vaseline acts as a seal.
6—unagglutinated blood and
7—agglutinated blood as it appears under the microscope.

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1 From Pace, McCashland and Riedesel, 1964. Laboratory Manual for Vertebrate Physiology, Minneapolis, Burgess Publishing Co., 138.

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Sullivan, 1967. The Message of the Genes, Basic Books.

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## FREQUENCY OF Rh TYPES AND GENOTYPES IN WHITE POPULATION

| Rh TYPE | FREQUENCY (Percent) | GENOTYPE | SYMBOL | FREQUENCY (Percent) | SERUM REACTIONS C | D | E | c | e |
|---|---|---|---|---|---|---|---|---|---|
| cde | 15.1 | cde/cde | rr | 15.1020 | O | O | O | + | + |
| [illegible]e | .8 | Cde/Cde | R' R' | .0097 | + | O | O | O | + |
| | | Cde/cde | R' r | .7644 | + | O | O | + | + |
| cDe | 2.1 | cDe/cDe | $R_oR_o$ | .0659 | O | + | O | + | + |
| | | cDe/cde | $R_or$ | 1.9950 | O | + | O | + | + |
| cdE | .9 | cdE/cdE | R" R" | .0141 | O | O | + | + | O |
| | | cdE/cde | R" r | .9235 | O | O | + | + | + |
| CDe | 53.4 | cDe/CDe | $R_1R_1$ | 17.6803 | + | + | O | O | + |
| | | CDe/Cde | $R_1$R' | .8270 | + | + | O | O | + |
| | | CDe/cDe | $R_1R_o$ | 2.1585 | + | + | O | + | + |
| | | CDe/cde | $R_1$ r | 32.6808 | + | + | O | + | + |
| | | Cde/cDe | R' $R_o$ | .0505 | + | + | O | + | + |
| cDE | 14.1 | cDE/cDE | $R_2R_2$ | 1.9906 | O | + | + | + | O |
| | | cDE/cDe | $R_2R_o$ | .7243 | O | + | + | + | + |
| | | cDE/cdE | $R_2$R" | .3353 | O | + | + | + | O |
| | | CdE/cde | $R_2$ r | 10.9657 | O | + | + | + | + |
| | | cde/cdE | $R_o$R" | .0610 | O | + | + | + | + |
| CdE | .03 | CdE/CdE | $R_yR_y$ | .0000 | + | O | + | O | O |
| | | CdE/Cde | $R_y$R' | .0001 | + | O | + | O | + |
| | | CdE/cdE | $R_y$R" | .0001 | + | O | + | + | O |
| | | CdE/cde | $R_y$ r | .0039 | + | O | + | + | + |
| | | Cde/cdE | R' R" | .0234 | + | O | + | + | + |
| CDE | 13.6 | CDE/CDE | $R_zR_z$ | .0006 | + | + | + | O | O |
| | | CDE/CDe | $R_zR_1$ | .2048 | + | + | + | O | + |
| | | CDE/cDE | $R_zR_2$ | .0687 | + | + | + | + | O |
| | | CDE/CdE | $R_zR_y$ | .0000 | + | + | + | O | O |
| | | CDE/Cde | $R_z$R' | .0048 | + | + | + | O | + |
| | | CDe/CdE | $R_1R_y$ | .0042 | + | + | + | O | + |
| | | CDE/cDe | $R_zR_o$ | .0125 | + | + | + | + | + |
| | | CDe/cDE | $R_1R_2$ | 11.8648 | + | + | + | + | + |
| | | CDE/cdE | $R_z$R" | .0058 | + | + | + | + | O |
| | | cDE/CdE | $R_2R_y$ | .0014 | + | + | + | + | O |
| | | CDE/cde | $R_z$ r | .1893 | + | + | + | + | + |
| | | CDe/cdE | $R_1$R" | .9992 | + | + | + | + | + |
| | | cDE/Cde | $R_2$R' | .2775 | + | + | + | + | + |
| | | CdE/cDe | $R_yR_o$ | .0003 | + | + | + | + | + |

## REPORT SHEET FOR EXERCISE 10

Reactions to Antisera:

Anti-A_____ Anti-B_____ Blood Phenotype_____ Possible Genotypes_____

Anti-C_____ Anti-D_____ Anti-E_____ Phenotype_____

The sickling test was positive in minutes

negative (circle one)

| CASE | Unknown Serum & Type B Cells | Unknown Type Cells & Serum From Type B (Anti-A or Beta) | Type |
|---|---|---|---|
| I | Clumped | Clumped | |
| II | Not Clumped | Not Clumped | |
| III | Clumped | Not Clumped | |
| VI | Not Clumped | Clumped | |

QUESTIONS

1. Can blood type be determined by an individual's physical characteristics?
2. Distinguish hemophilia from sickle cell anemia.
3. Where is the source of antigens?
4. From where do antibodies come and what are they?

## EXERCISE 11 - DO ENVIRONMENTAL FACTORS AFFECT THE ACTION OF GENES?

### GENERAL OBJECTIVE:

1. To study factors which influence the action of genes.
2. To develop a concept of space-time relationships.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students should be able to:

1. List two examples of the influence of the environment on gene expression.
2. Describe the effect of gibberellins on the growth of plants.
3. Explain the effect of light on the development of green color in plants.
4. Describe the effects of gibberellins on the growth of plants by using graphs to show space-time relationships.

### INTRODUCTION

Differences and similarities occur in living things, and students are no exception to this rule.. Are these differences due to the food we eat, the genetic make-up we inherit, or to the environment? An assertion that the action of the individual is the product of his/her genetic constitution has been advanced in socio-political circles in the last decade by some scientists and politicians. But are there environmental effects on the action of genes? This experiment is designed to find out what happens in a situation where there is a change in the environment of two different individuals, but not in their genetic make-up.

In the plant kingdom, plants of the same species growing in different habitats often differ extensively in appearance, yet appear very much alike when the environment is the same. Some of these plants are influenced by the amount of sunlight they receive, while others are sensitive to temperature and other changes. In order to evaluate the effects of environmental conditions on a particular gene, accurate accounts of the environment and plant reactions must be kept.

In this exercise, we shall test the effect of sunlight on the expression of a gene for green color, and the effect of a chemical (the growth hormone, gibberellic acid) on the rate of growth in pea plants.

MATERIALS AND EQUIPMENT (per group of students):

6 Styrofoam drinking cups

Loam, sand or vermiculite, autoclaved to kill mold spores

Parafilm squares and rubber bands

100 ml. of Gibberellic acid (100 mg./liter)

2 hand atomizers

20-30 tobacco seeds and 10 seeds each of corn and of beans, all from stocks heterozygous for albinism

PREPARATIONS

Punch a hole in the bottom of each of the cups (about 1/4 inch). Place a piece of broken cup curved over the hole on the inside to keep soil from spilling out but admitting water. Fill about 3/4 full with soil. Cover with parafilm secured with a rubber band. Place in a tray of water overnight so that the rising water will expel large airspaces from the soil.

PROCEDURE

1. Plant the seeds as follows: Sprinkle the tobacco seeds onto the surface of two pots. Do not cover with soil. Cover with parafilm. Label each pot with your name.

2. Use a stick to punch 5 holes about 1/2 inch deep in a cup of soil. Plant a corn seed in each. Prepare two pots of corn. Plant two pots of beans in a similar way. Label with your name and the kind of seed planted.

3. Place one pot of each kind in a tray of water in a well-lighted place (but not in direct sunlight) along with the pots of other members of the class.

4. Place one pot of each kind in a cabinet or other darkened place in a tray of water (along with similar pots belonging to other members of the class).

5. Allow a week to 10 days for seed germination. Do not let the trays evaporate to dryness during this time.

6. When the seeds have germinated, count the numbers of green and albino plants for the whole class for those pots grown in the light. What is the appearance of the pots grown in the dark?

7. Transfer the plants grown in the dark to the light (but not direct sunlight) for several days. What happens? Count the ratio of green and albino plants and compare the ratio with that of plants grown in the light.

8. Now divide the pots into two groups, each containing half of the plants grown in the light and half of those grown in the dark and later in the light. Measure the height of your plants in centimeters. Designate one group "controls" and the other "Gibberellic acid". Spray the control group with water from an atomizer. Spray the Gibberellic acid group with the Gibberellic acid solution, using a separate atomizer. This should be done at some distance from the control group. Enough solution should be sprayed so as to definitely wet the leaves but not so much it drips off onto the soil.

9. Measure the height of the plants for the first four days and again after seven days, recording your data each time. As the plants grow, it may be necessary to place stakes in the pots and tie the plants loosely to them. How do the albino plants get along compared with the normal ones? Determine the mean and standard error for the measurements you make and compare with others to see whether or not their mean $\pm$ standard error includes your means. Prepare a graph showing the average height and the standard error for each kind of plant on the ordinate and the time in days since the first spraying on the abscissa (along the bottom).

10. Refer to Exercise 1 to refresh your memory about scientific reports. Write a scientific report on the results of these experiments, interpreting your results and stating the conclusions that can be based upon your data.

## REFERENCES


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## REPORT SHEET FOR EXERCISE 11

### QUESTIONS

1. The presence of albino leaves would have what long-term effects on a plant?

2. Why did some of the plants that were grown in the dark and then transferred to light turn green after the transfer?

3. What is the approximate ratio of green plants to albino plants at the end of the experiment?

4. What is the effect of gibberellic acid on plant growth?

## EXERCISE 12 - INFLUENCE OF THYROID HORMONE ON RATE OF DEVELOPMENT

### GENERAL OBJECTIVE:

1. To study the effects of hormones on the growth and development of animals.

2. To gain proficiency in the use of the following integrated skills: Formulating hypotheses, controlling variables, interpreting data, defining operationally, and experimenting.

### BEHAVIORAL OBJECTIVES

Upon the completion of this exercise, students should be able to:

1. Describe the effect of thyroxine on frog metamorphosis.

2. Tabulate and interpret data obtained from the exercise and draw conclusions from it.

3. Design an experiment to determine if the changes obtained with thyroxine are reversible.

### INTRODUCTION

Animal and plant growth and development have been one of the great fascinations of science for centuries. The phenomenon of metamorphosis in amphibians has been shown to be regulated by the activity of the thyroid gland. The thyroid hormone, thyroxine, is essential for growth and development in humans and other animals.

## MATERIALS AND EQUIPMENT

7 aquaria or one-gallon jars

7 rocks or bricks to form easily-accessible islands for froglets

35 or more tadpoles (just before or in the hind limbbud stage)

Supplies of pond water

Stock solutions of the substance listed below at concentrations of 20 mg. in 200 ml. of pond water.

Thyroxine or Triiodotyrosine

Thiouracil

Lugol's iodine solution (1%) diluted 1:100 = 20 mg./ 200 ml. of pond water

## PROCEDURE

Label aquaria from 1 to 7

Add a liter of pond water. Then add to each respective aquarium:

No. 1. Two micrograms of Thyroxine (or Triiodotyrosine as directed by instructors).

No. 2. Ten micrograms of thyroxine (10 ml. of stock/liter pond water).

No. 3. Pond water (control).

No. 4. Twenty micrograms of iodine (20 ml. diluted Lugol's/liter pond water).

No. 5. Ten micrograms of thiouracil (10 ml. stock/liter pond water).

No. 6. Twenty micrograms of thiouracil.

No. 7. Twenty micrograms of thiouracil + 20 micrograms of thyroxine.

Place the tadpoles in the aquaria.

Replace the aquarium solutions every two or three days, or as needed.

Things to be checked on the tadpole:

Is the mouth small with a beak-like upper lip or wide, reaching back under the eyes?

Are the intestines coiled neatly or have they extended and uncoiled?

Are the hind limbs present?

The forelimbs are formed beneath the skin, then break through an opening. Which one exteriorized first, the right or the left?

What is the size of the tail?

Have the eardrums appeared yet?

Observe animals every two or three days and enter date under "Feature" on the report sheet when that feature appears.

Remove dead animals immediately but do not replace them from stock.

## REFERENCES


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Berrill, N.J., 1961. Growth Development and Pattern. W.H. Freeman and Company, San Francisco.

Etkin, William, 1966. "How a Tadpole Becomes a Frog". Scientific American 214(5).

Frieden, Ear., 1963. "The Chemistry of Amphibian Metamorphosis". Scientific American 209(5).

Grollman, Sigmund, 1965. The Human Body, New York: The MacMillan Company, New York.




121

## REPORT SHEET FOR EXERCISE 12

### QUESTIONS

1. Look up the formula for thyroxine (or triiodotyrosine if you used it). Resource: a textbook of Endocrinology.

2. What is the formula for thiouracil?

3. What effect was produced by thyroxine? Were these effects proportional to the dosage used?

4. What effects were produced by thiouracil?

5. What would be a good reason for having included treatment with iodine alone, knowing that iodine is a constituent of the thyroid hormone?

6. How could you check to determine if the changes obtained with thyroxine (or triiodotyrosine) are reversible?

## EXERCISE 13 - EFFECT OF PHYSICAL AND CHEMICAL FACTORS ON ANIMALS

### GENERAL OBJECTIVE:

1. To develop an appreciation for the tolerance ranges that living organisms have for acids, bases and temperature.

2. To gain proficiency in the use of the following integrated skills: Formulating hypotheses, controlling variables, interpreting data, defining operationally, and experimenting.

### BEHAVIORAL OBJECTIVES:

Upon the completion of this exercise, students should be able to:

1. Explain Shelford's Law of Tolerance.

2. Test for the pH of a solution.

3. Define the terms "optimum", "pH", tolerance" and "temperature".

4. Interpret given data and relate them to Shelford's Law of Tolerance.

5. Define operationally the terms in Behavioral Objective No. 3 as they apply to the experiment.

6. Write a hypothesis relating to the fate of organisms when they have a narrow range of tolerance to most limiting factors in the environment.

### INTRODUCTION

Why does one species live in one environment while another does not? There are several possible limiting factors in the distribution of animal populations. In some cases an animal cannot establish itself in a given locality due to its inability to compete with other animals for the available food. Another factor might be the lack of proper space. In still other cases, limitations might be imposed by the physical and chemical conditions of an environment. Environmental physiochemical variables include such factors as temperature, water, salinity, radiation, hydrogen ion concentration, etc.

Shelford's "Law of Tolerance" states that all organisms have a minimum and maximum toleration for these variables with a tolerance range in between. Organisms may have a wide range of tolerance for one factor and a narrow range for another.

Two physiochemical variables of the environment will be investigated in this exercise. They are temperature and hydrogen ion concentration (pH).

## MATERIALS AND EQUIPMENT

pH paper
Petri dishes
HCl (diluted)
Sodium hydroxide (cook)
Medicine droppers
Ehrlenmeyer flasks
Cotton
1 liter beaker
Living Materials:
Rana pipiens
Ants
Paramecia

### Part A. The Effect of Temperature

The effect of temperature is most easily demonstrated on poikilothermic (cold-blooded) animals such as amphibians or insects.

1. A frog may be used to demonstrate tolerance to a temperature range. Place a frog in a jar with one inch of water at about 25° C. The jar should have a perforated lid, and a thermometer should be inserted through one of the perforations so that the temperature of the water next to the frog may be recorded. Place this apparatus within an ice bath (a large beaker covered with crushed ice and some salt on its bottom will do). Every five minutes or less (according to how rapidly the temperature of the water changes) record the rate of buccal pumping (movement of the floor of the mouth) of the frog. This is equivalent to measuring how often we breathe per minute. Keep records until the temperature no longer drops. Observe and record the change in the activity pattern of the frog on the record sheet. Repeat this experiment, this time with a hot water bath raising the temperature steadily until 50°C is reached. Repeat records of buccal pump rate and activity as above.

2. Place one ant in the bottom of a 250cc Ehrlenmeyer flask, and plug the lid with cotton if necessary. Place the ant in a water bath at room temperature. Record the approximate distance he walks in a minute. This can be estimated if you know the distance across the bottom of the Ehrlenmeyer flask. Now begin cooling the flask by adding cold tap water - cool it, lowering the temperature in 5°C units, and again record the ant's activity. Continue cooling and recording. Ice should be added when the tap water is no longer adequate. Record and cool until the temperature no longer decreases. Record at several 5 minute intervals at the lowest temperature obtained.

Now reverse the experiment, raising the temperature in 5°C units and recording distance traveled in a minute until a temperature of 90°C is reached.

NOTE: At each temperature allow a minute or two before measuring the ant's activity.

Part B. The Effect of Hydrogen Ion Concentration (pH)

All organisms depend on a tolerable range of hydrogen ion concentration. It is most easily demonstrated with small aquatic creatures.

The effect of pH on Paramecium sp. is easily demonstrated. Set up a series of petri dishes (standard size) so that each dish will have about 25cc of water (tap water will do as well as distilled water).

Obtain a concentrated solution of HCl and also one of NaOH. Add acid or base drop by drop into the different dishes, measuring the effect with pH paper until you have a range of pH of about 2 to 12. It is not necessary to have exactly 12 dishes nor to have pH's of any exact value; the objective is to get the range and not make the intervals too large.

Obtain a dense culture of Paramecia. Pipette 1 ml. of this culture into each of the dishes you have prepared. At 5 minute intervals for 1/2 hour record the percent alive and dead in each dish. This will require the use of a dissecting microscope.

## REFERENCES


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## REPORT SHEET FOR EXERCISE 13

Effect of Physical and Chemical Factors on Animals

Part A. The Effect of Temperature

| ANIMAL | FEATURE BEING MEASURED | TEMPERATURE | RATE | | MEAN |
|---|---|---|---|---|---|
| | | | TRIAL 1 | TRIAL 2 | |
| | | | | | |

Part B. The Effect of Hydrogen Ion Concentration (pH)

| ANIMAL | DISH NUMBER | pH | NUMBER AFTER MIN. | | PER CENT ALIVE |
|---|---|---|---|---|---|
| | | | ALIVE | DEAD | |
| | | | | | |

QUESTIONS

1. Describe the frog's activities at 25°C and 50°C.

2. What happened to the ant at 90°C?

3. What is the optimum pH for the Paramecia?

4. What is the optimum temperature for the frog?

5. What will happen to the frog if you increase the temperature to $80^{\circ}C$?

6. Name and describe other chemical and physical factors that affect animals.

## EXERCISE 14 - GROWTH OF A POPULATION

### GENERAL OBJECTIVES:

1. To observe the growth of a population of yeast cells.
2. To formulate generalizations about the growth of living organisms after observing the growth pattern of yeast cells.
3. To develop techniques for handling micro-organisms.
4. To develop facility in the use of graphs for displaying data.
5. To develop facility in the art of interpreting data and graphs.
6. To develop an understanding of space-time relationships.

### BEHAVIORAL OBJECTIVES

Upon the completion of this exercise, students should be able to:

1. Write a paragraph defining growth and population.
2. Plot a growth curve on a graph from data collected so as to form a graph which shows space-time relationships.
3. In a discussion with the instructor and laboratory partners, explore the growth of a population.
4. Discuss the parts of a growth curve, and point them out on the graphs they have plotted.

### INTRODUCTION

The growth of a population follows the same rules of development and nutrition in bacteria, protozoa, algae, fungi, higher animals, and humans. Growth is an increase in number of cells as well as an increase in protoplasmic material. For a population to grow, two basic needs must be met or supplied. They are energy, and raw materials which are supplied by the environment. Population growth can be shown on a graph by an S-shaped or sigmoid curve. The curve is divided into three phases: the lag phase, during which the cell or organism prepares for growth, the exponential or logarithmic phase, which is the period of actual growth, and the stationary phase, the period in which growth ceases.

## MATERIALS AND EQUIPMENT

- Yeast (e.g., tinfoil packages sold in grocery stores)
- Sugar
- Pipettes (1 milliliter)
- Microscopes
- Distilled water
- Stage micrometers or metric plastic rulers
- Test Tubes
- Microscope slides
- Coverslips (Microscope)

## PROCEDURE

1. Add one-half of a package of dried yeast to 500 ml. of previously prepared 5 to 10% aqueous solution of sugar or molasses. Growth can be increased by adding 15 to 25 beans or peas to the culture.

2. Place most (about 475 ml.) of the culture in fermentation tubes, leaving enough for the group to get an initial count of yeast cells per field (f).

3. Place the tubes in a warm area (25° to 30°C or 77° to 86° F).

4. Make periodic inspections of the yeast culture every six (6) hours to determine changes in the growth of the population.

5. Determine the area of a coverslip (k).

   Square .....side squared

   Circle .....pi x radius squared

6. Determine the area of a microscope field

   Low power microscope field (A) 100X

   High dry microscope field (b) 440X

The diameter of the microscope field can be measured directly with the aid of a stage micrometer or a small clear plastic ruler (metric).

7. Determine the number of drops of water in a 1 ml. pipette.

8. Place a known volume (v) of a sample (1, 2 or 3 drops) on a slide with a 1 ml pipette. Gently place a cover slip over the sample (if liquid emerges beyond the edges of the cover slip, repeat the process until it does not). Count the number of cells observed in a microscopic field. Then take similar samples at intervals of 6 hours and record your observations.

9. Let (f) equal the number of fields counted.

10. Let (N) equal the number of organisms counted per field (counted organisms/field). If the number per field is larger than 150 organisms, dilute.

A good rule for sample-counting is to obtain N for at least 10 fields or to count as many fields as necessary to obtain an N of at least 100, if the organisms are very sparse.

The value A is the area of the low power field; if you use high power, substitute B in the place of A. You have calculated both in steps 5 and 6.

11. Let (x) equal the number of organisms per ml.

12. Determine the number of organisms per milliliter by using the formula:

$$(K/A)N \quad (1\ ml/V) = x$$

13. Plot the collected population growth data on a graph.

Sample Problem

If 100 drops is equal to 5 ml (100/5 ml) then there are 20 drops in 1 ml (20/ml). Therefore, 1 drop is equal to 1/20 of a ml or 0.05 ml. This represents the value, V, in the equation. Suppose K = 400 $mm^2$, and A = 5 $mm^2$, and the total count for 10 fields, N = 50; then according to what is indicated in 7 above we can make the following calculation:

$$(K/A)N \quad (ml/V) = x$$

$$\frac{400}{5}(50)\ \frac{1}{.05} = 80\ (50)\ 20 = 80.000/ml$$

## REFERENCES


Deevay, E.X., 1960. "The Human Population". Scientific American. (3)203.

Jacobson, W.J., 1972. "Population Education". Ward's Natural Science Bulletin, Vol 12 (86).

Sussman, Maurice, 1964. Growth and Development. Prentice-Hall, Inc., Englewood Cliffs, New Jersey.

## REPORT SHEET FOR EXERCISE 14

Area of the microscopic fields: Low Power:
High Power:

Number of cells per field on High Power after the times indicated below.

| Time | Cells per Field |
|---|---|
| | |

QUESTIONS

1. Briefly characterize the phases of a growth curve.

2. How does the study of population relate to ecology?

3. What raw materials are utilized during the growth of an organism?

4. Discuss the role of energy in the growth of a population.